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> *"To the solid ground*
> *Of nature trusts the Mind that builds for aye"*—Wordsworth

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Saturday, July 16, 1960

DISPOSAL OF RADIOACTIVE WASTE

THE problem of waste disposal is as old as life itself, and although it has raised difficulties in the past, there is no reason why the disposal of radioactive wastes should not be carried out in a completely satisfactory manner. Man's knowledge of the deleterious effects of radiation is by no means complete, but it is probably as extensive as his knowledge of any other single potential source of injury. Moreover, his attitude towards radiation is one of greater concern and caution than towards any other comparable danger. At the same time, almost all users of radioactive material recognize that they will damage the future of their industry, perhaps irreparably, if they adopt an irresponsible attitude towards the disposal of radioactive waste. In return, they have the right to expect a balanced appreciation and understanding of their problems by the community for which they work.

The importance of these problems was emphasized by the Conference on Radioactive Waste Disposal organized at Monaco in November by the International Atomic Energy Agency and the United Nations Educational, Scientific and Cultural Organization. This Conference, with about three hundred participants from more than thirty countries, dealt with the handling and treatment of radioactive waste as well as its disposal into the atmosphere, the sea and the land. The Conference programme gave special prominence to the marine problems, partly because the Conference was held in the Oceanographic Museum of Monaco, but mainly because there is at present a widespread international interest in the freedom of the seas and the utilization of marine resources.

The problems of waste disposal involve many scientific disciplines, and one of the principal aims of this Conference was to draw these disciplines together. In this it has succeeded to a considerable extent. However, too much time was spent in discussing the treatment and handling of waste prior to disposal, and there was no attempt to explain the technicalities of radioactivity to specialists in other fields, many of them clearly meeting these technicalities for the first time. There was also some repetition of information on the types and quantities of wastes from national atomic energy projects, and this repetition could have been avoided by the presentation of a single paper reviewing the problems from an international point of view. In spite of these weaknesses, considerable progress was made towards explaining the problems of the user or producer of radioactivity, and conversely the representatives of atomic energy projects now have a better understanding both of the natural fears of the biologist and ecologist and of the extent to which these and other specialists can help them in achieving the safe disposal of waste.

Technically, this was not a conference of disclosures, and probably most of the information presented was already known to specialists in the relevant fields. The progress which has been made has been in the field of defining principles and how they should be applied, and understandably this has not been achieved without differences of opinion, some of which have not yet been resolved. These differences did not appear during discussions on gaseous waste, nor in sessions dealing with disposal into geological formations. They were almost entirely confined to the problems of the marine environment. The two subjects which aroused the greatest interest were the discharges of low and intermediate activity wastes to the sea from Windscale, in Cumberland, and the technical and legal problems posed by the international nature of the sea. Although some of the questions contained critical implications, there was little direct criticism of the Windscale discharges, and a favourable impression was created by the reports of the considerable amount of work undertaken before and during the discharges. There was, however, a complete opposition of views on the legal question. It was the general view that present international law is not adequate to deal with radioactive waste, and that eventually it will be necessary to agree on an international convention. A strongly expressed minority view considered that all discharges of radioactivity to the sea are dangerous and that it would be wrong to discuss any proposals for legalizing them.

The British view at the Conference, supported by American oceanographers and biologists, was that the use of radioactive materials and nuclear power necessarily involves the releases of small amounts of radioactivity to man's environment. These releases cause some additional radiation exposure of man, and their significance can be judged by comparing this exposure with the recommendations of the International Commission on Radiological Protection. The exposures must be kept below the recommendations of that Commission regardless of the cost in human effort, and it is desirable to keep them as far below these recommendations as is practicable. In assessing whether a reduction is practicable, it is necessary to consider the level of exposure as a fraction of the Commission's recommendations and the reduction factor likely to be achieved, as well as the effort involved in achieving it. This view appears to be shared by the representatives of almost all the atomic energy projects. An alternative view which appeared to underlie some of the contributions, and which was expressed in more detail in personal discussions, holds that no expenditure of effort is too great if it reduces human exposure, and that, with sufficient effort, releases of radioactivity to man's environment can be reduced to zero. In its simplest form, this view was expressed as an outright condemnation of any discharges of radioactivity to the sea, or to other parts of man's environment. The weakness of the position adopted by the supporters of this view is in the absolute nature of their claim. Some quantities of radioactivity are demonstrably trivial in any environment, and a complete denial of this can have only one practicable meaning—that no use can be made of radioactive materials or nuclear power. Clearly the supporters of this absolute point of view are not anxious to achieve the banning of artificial radioactivity and nuclear power; their aim is presumably that of all the specialists in this field, namely, to deal with radioactive waste in ways which do not harm man or spoil his environment. By refusing to admit the possibility of agreeing either on scientifically based standards or on legal provisions controlling the disposal of waste at sea, they are actually reducing the chances of achieving this aim.

It would be wrong to under-estimate the importance of these differences of opinion, and we shall certainly hear more of them in future conferences on atomic energy and on such things as the law of the sea. On the other hand, they were by no means a dominant feature of the Monaco Conference, which should be remembered more for the opportunities it gave for exchanging views. There was complete agreement that nuclear power must not be allowed to impair man's use of the resources of the sea, either now or in the future, but as one prominent oceanographer remarked—the ability to receive wastes without damage *is* one of the resources of the sea. If an appropriate balance is to be obtained between the utilization of marine resources and the cost to mankind of nuclear power, the oceanographers and marine biologists must join the chemists and chemical engineers in quantitative studies aimed at providing governments with the scientific basis of a balanced decision.

BIOLOGICAL CONCEPTS BEFORE DARWIN

Forerunners of Darwin

1745–1859. Edited by Bentley Glass, Oswei Temkin and William L. Straus, Jr. Pp. viii+471. (Baltimore, Md.: The Johns Hopkins Press; London: Oxford University Press, 1959.) 52*s*. net.

WHEN a man has done many things he is more likely to have had numerous forerunners than if he had done nothing, and their number is no disparagement to his achievement, as can be seen in the case of Darwin. This is all the more so when the forerunners are selected from only a part of his fields of activity, represented by evolution by natural selection and omitting his work on coral reefs, volcanic islands, insectivorous plants, climbing plants, cross-fertilization, and methods of communication between animals by means of the expression of the emotions. The forerunners in the field of transmutation of species and the methods by which it has been brought about are by themselves quite sufficient to fill a splendid volume.

From a recently discovered letter addressed by Darwin to Baden Powell, the contribution which Darwin claimed as his own unaided achievement is now precisely known. It is to have discovered the explanation of "*how* species become modified, & to a certain extent how the theory of descent explains certain large classes of facts", in which respect he received no assistance from his predecessors. As more and more manuscripts come to light, and existing documents are studied more critically, the number of forerunners in any field increases. With the help of hindsight, as Prof. Loren C. Eiseley has shown, it is possible to read into some works certain ideas which, in fact, their authors did not consciously recognize. He has established a criterion which must be satisfied before a forerunner of Darwin can be accepted as a real precursor. There must be recognition of the similarity between artificial and natural selection, and the conception of unlimited organic change in vast periods of time. Played according to these rules, the exploration of biological concepts prior to Darwin is a fascinating game to which the book under review is a perfect guide. Its fifteen chapters are as many essays on various aspects of the theory by a number of recognized experts, and the fact that some points here and there will lead to disagreement on matters of detail only shows how stimulating and thought-provoking the book is.

GAVIN DE BEER

PEDOLOGY

Fundamentals of Soil Science

By Prof. C. E. Millar, L. M. Turk and Prof. H. D Foth. Third edition. Pp. x+526. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1958.) 62*s*. net.

Introduction to Soil Science

By Prof. G. W. Leeper. Third edition. Pp. viii+222+12 plates. (Carlton: Melbourne University Press; London: Cambridge University Press, 1957.) 30*s*. net.

The Study of the Soil in the Field

By G. R. Clarke. Fourth edition. Pp. 204+6 plates. (Oxford: Clarendon Press; London: Oxford University Press, 1957.) 35*s*. net.

ALTHOUGH pedology has now advanced to the point where it has become a coherent discipline worthy to be taught as a subject in its own right at university-level, it has for too long been regarded simply as a branch of husbandry, taught in agricultural faculties for agricultural ends. One result of this is the present lack of adequately trained pedologists; another is the virtual absence of good up-to-date text-books which develop the whole subject in a balanced and logical fashion. Unfortunately, neither of the two general text-books under review does anything to rectify this situation

"Fundamentals of Soil Science" in no way lives up to its title. It is a superficial descriptive text, adequate, perhaps, for potential farmers familiar with North American conditions but quite unsuitable as the university text-book which it claims to be, for two main reasons.

In the first place, it fails to take advantage of the results of modern work (for example, on the structures and colloid chemistry of the silicate minerals) to explain in fundamental terms the processes occurring in soils. Secondly, it glosses over all debatable points, making no attempt to discuss them, and the inquiring student will find no bibliography. The book, therefore, gives neither a sound training in pedology nor any encouragement to the student to use his powers of reasoning. Instead, it branches off into the realms of crop husbandry and a very long way from its own title. Further, the book is too long and too expensive for the material it contains, and the reader is irritated by the repetition of section headings in the form of little questions in the text. The inclusion of some good diagrams and photographs and a useful glossary unfortunately do not affect the criticisms given above.

"Introduction to Soil Science" is a far less ambitious book and is considerably shorter and cheaper. It is aimed at a similar audience but with Australian conditions especially in mind. The text is written in a clear, concise style and is readable and interesting. As the author himself points out, the arrangement of the chapters is not as logical as it might be—soil erosion, for example, is considered in the section on soil chemistry rather than with soil physics. Some suggestions for practical work are given in the appendix but, as with the material of the book as a whole, these experiments fall between the requirements of the serious university student and those of the schoolboy who may be interested in soils. There is a short bibliography, mainly applicable to Australian conditions.

"The Study of the Soil in the Field" is a very different proposition. This book, now in its fourth edition, represents the accumulated wisdom of one who has studied soils in the field for many years and who is something of a philosopher. It is primarily a lucid and concise text-book of field procedures and is virtually indispensable for anyone, be he student or practising pedologist, who wishes to study the soil in its natural context. The book is essentially written in the light of experience in Britain, but frequent comparisons are made with conditions elsewhere. A new addition is the chapter on the use of air photographs in soil surveys. It contains useful information on air photographs in general, but becomes rather vague in the section on actual interpretation for soil survey purposes. The photographs with accompanying traces are very interesting, but demonstrate the usefulness of air survey for geological and ecological rather than for pedological purposes.

R. M. S. Perrin

PRACTICAL INSECT MORPHOLOGY

Manual of Insect Morphology

By Prof. E. Melville DuPorte. Pp. xi+224. (New York: Reinhold Publishing Corporation; London: Chapman and Hall, Ltd., 1959.) 40*s*. net.

This volume is a publication of the typescript notes used by the author when he was teaching his laboratory class. It gives direct instructions for making nearly two hundred examinations and dissections, using insects that can easily be obtained in quantity. Naturally, North American insects are specified, but equivalent substitutes can be found in other regions. After familiarizing the student with the idea of segmentation, and with the basic construction of a segment, the manual gives ten chapters to external structures, and six to internal organs.

Instructions are explicit, and the language is commendably simple and direct. New terms are put in italics the first time that they are introduced, but in spite of this a student would fairly often come upon a word that is not defined in the text nor in the inadequate glossary on p. 6. He would have to remember such words as 'condyle' and 'invagination' from the lectures that he is presumably attending concurrently with his laboratory course.

So far as the text goes, this manual is excellent. It is clearly printed, helpfully divided into convenient sections, and uses a variety of type-faces to the best advantage. It deserves high praise for presenting so much information in such a readily accessible form.

The problem for the reviewer, and for the potential user, is to know what to say about the almost total lack of illustration. There are only fourteen figures, all schematic line-drawings of the simplest kind. We are told that they are intended as a guide to the structure, and that none of them is an exact reproduction of any known insect. In fact, they are the sort of diagram that a lecturer might rough out on the blackboard, talking and explaining all the while; without the lecturer to explain them they do not add much to the text. There are no drawings at all of what one may expect to see after making any of the dissections. "Students are advised to study the drawings in the text-book or other writings of Snodgrass, as well as the work of other competent entomological illustrators." One can sympathize with the author's problem. Two hundred anatomical drawings, fully lettered, would have cost a lot to make, and more to publish. The price of this book is already too high for what it is, and with such figures it would be prohibitive. Yet without any figures I do not see how it can be used by an unaided student. He has neither the time nor the experience needed to be able to apply a verbal description, however lucid, to an unskilled dissection.

The manual may be useful to some experienced entomologists—a demonstrator called upon to give a course in insect morphology, or a specialist in one group who wishes to study the anatomy of related groups.

H. Oldroyd

ULTRASONIC PROPAGATION

Absorption and Dispersion of Ultrasonic Waves

By Karl F. Herzfeld and Theodore A. Litovitz. (Pure and Applied Physics: a Series of Monographs and Text-books, Vol. 7.) Pp. xviii+535. (New York: Academic Press, Inc.; London: Academic Press, Inc. (London), Ltd, 1959.) 14.50 dollars.

Since the appearance of the paper of P. Debye on the effects of high-frequency relaxation in the propagation of electro-magnetic waves in substances, much research has been done in that field of study and many books and papers published. Less attention has been devoted to the corresponding relaxation effects in the propagation of compressional waves, though the discovery by G. W. Pierce in 1925 of a dispersion in the velocity of high-frequency sound waves in carbon dioxide gas marked the beginning of new studies which have occupied a considerable number of research workers in physics and physical chemistry since.

It is opportune that we should now have a general survey of acoustical relaxation effects; and the authors of this volume, by their researches on both the theoretical and experimental aspects, are well equipped to make it. The plan of the book is succinctly set forth in the introduction. The evidence for relaxation, shown by a frequency-dependent specific heat and consequent excess absorption, is examined in the light of various theories, in particular those based on the idea of a bulk viscosity. From single relaxation times we pass to multiple ones and to interactions between different types of molecules in mixtures.

The weakness of all theories in this field has been that, though the existence of relaxations had been experimentally proved, the relaxation times themselves could not be deduced *a priori*, but in Chapter 7 the probability of energy transfer during the passage of the sound wave is discussed, and calculations made of the ratio of the relaxation time to the mean free time between collisions for a diatomic gas, and hence of the relaxation time itself as a function of temperature.

Later the experimental results for liquids are presented. The relevant theory is much more difficult, and at present one can only discuss the results phenomenologically.

The book is an excellent and detailed account of the present state of ultrasonic propagation in fluids and should be read by all who work in this field or wish to examine the latest theories of relaxation in fluids. There is no discussion of ultrasonic propagation in solids and, in this respect, the title is too general; although, as the authors point out, there exist books which cover the omission.

E. G. RICHARDSON

NUCLEAR POWER TECHNOLOGY

Nuclear Engineering Handbook

Edited by Harold Etherington. (McGraw-Hill Handbooks.) Pp. xv + 1866. (London: McGraw-Hill Publishing Company, Ltd., 1958.) 194*s*.

Nuclear Power Plant

By E. Openshaw Taylor. Pp. vii+184. (London: George Newnes, Ltd., 1959.) 30*s*. net.

Chemistry of Nuclear Power

By Dr. J. K. Dawson and Dr. G. Long. Pp. vi+208+8 plates. (London: George Newnes, Ltd., 1959.) 30*s*. net.

Physics and Heat Technology of Reactors

Translated from the Russian. (Supplement No. 1 of the Soviet Journal of Atomic Energy, *Atomnaia Energiia*.) Pp. v+174. (New York: Consultants Bureau, Inc.; London: Chapman and Hall, Ltd., 1958.) 22.50 dollars; 168*s*. net.

THE "Nuclear Engineering Handbook" is an attempt to collect into one volume theory, formulæ and data from the many different branches of science and engineering which have contributed to the development of nuclear fission reactors. The contents include sections on mathematics, nuclear and reactor physics, isotope technology, radiological protection, heat transfer, materials, chemistry and chemical engineering, and mechanical design. The editors have realized how important it is that a handbook should be easy to use and have provided, in addition to an index, an extensive guide.

The handbook contains an immense amount of information, extremely well organized. The kind of information given and the aim in giving it varies considerably from section to section. In the sections on mathematics the reader will find a neat review of algebra, geometry and analysis, starting from the basic arithmetical operations and including the theory of equations, vector analysis, probability, differential and integral equations, integral transforms, the calculus of variations, etc. In spite of its clarity, however, this section is not likely to satisfy either mathematicians or others seeking mathematical assistance in solving practical problems.

In the numerous sections dealing with what might be termed the nucleonics of reactors, a comprehensive treatment is given, starting with nuclear physics and leading to a comparison of reactor design formulæ with experimental results.

In the sections on fluid flow, heat transfer and materials, emphasis is placed on the application of information. The fundamentals of fluid flow are sketchily treated, but a large amount of useful data is included in the form of tables, graphs and equations. The designer is also catered for in a section which includes general and detailed drawings of several reactors and components.

In a book of this kind it is not easy to strike a balance between data and text. I would prefer greater emphasis on the former. In spite of these minor criticisms, the book is invaluable to anyone who needs to be constantly in touch with several of the diverse subjects involved in nuclear engineering.

"Nuclear Power Plant" contains an elementary treatment of the principles and problems of nuclear power plant design. The author has assumed that the reader's scientific standard is that of the sixth-form science courses in schools in Britain, and except in relatively unimportant respects he has successfully kept the requirements to this standard. The arrangement is logical, starting with a review of energy requirements and resources, continuing with the scientific and engineering aspects and concluding with a chapter on the economics of nuclear power stations. The important topics both in science and engineering have been well selected, and, although it is inevitable that simplifications are made in a book of this length, they are, in the present case, few and unimportant.

"Chemistry of Nuclear Power" describes the many important roles which chemistry takes in nuclear power technology. Although not written specially for chemists, an elementary knowledge of chemistry is essential to understand some sections. The part which chemistry plays in refining raw materials, in fuel preparation and re-processing, in reactor technology and in disposing of radioactive wastes is clearly indicated, as is the complicated relationship between chemistry and metallurgy. This book should go far to convince a sixth-form or first-year university student that the career of the chemist in nuclear technology is as interesting and important as that of the physicist.

"Physics and Heat Technology of Reactors" contains English translations of nineteen original papers by Soviet authors. The papers on reactor theory will be of special interest to Western readers because of the originality of the Soviet scientist in this field. Those dealing with heat technology are disappointing when seen in relation to the great amount of high-quality research in this subject which Soviet technologists have carried out.

W. MURGATROYD

PROFESSIONAL ENGINEERS IN BRITAIN

THAT engineering can be practised as a profession has been recognized in Great Britain for well over a century. The registration of professional engineers, a practice which has become established in a good many other parts of the world, has not found favour in Great Britain, and the standards of the profession have been built up and are maintained by the professional engineering institutions, notably the Institutions of Civil Engineers, of Mechanical Engineers and of Electrical Engineers.

There has been since the Second World War in all the engineering institutions a good deal of reviewing of academic standards, of requirements for practical training, and indeed in quite a fundamental sense of the meaning to be ascribed to the term 'professional engineer'. It is not therefore surprising that Sir Willis Jackson should choose as the theme of his presidential address to the Institution of Electrical Engineers, delivered on October 9, "The Making of Professional Engineers".

Education, training and the social and economic setting in which engineers pursue their vocation come under review in a balanced assessment and penetrating analysis of what might be termed the production and utilization of professional engineers in Great Britain. That the sustained effort which has been made in recent years to attract an increased proportion of the more able young people to take up careers in science or technology is beginning to bear fruit is clearly evidenced by data given in the address. The numbers of schoolboys and schoolgirls passing the Advanced Level General Certificate of Education examination in the subjects of physics, chemistry and mathematics increased between 1953 and 1958 by some 59 per cent, 43 per cent, and 65 per cent, respectively, the corresponding increase in school population over fifteen years of age in the period being some 26 per cent. While radio, television, the national Press and official publications have all shared in this effort, there is no doubt that a major contribution has been made by industry by fostering a closer contact between schoolmasters and the staff of industrial organizations. In addition, the teaching of experimental science in the independent schools has been stimulated and greatly assisted by the Industrial Fund for the Advancement of Scientific Education which was established in 1955.

Gratifying as this increase in numbers is, there is, on the other hand, little cause for satisfaction as to progress in the attempt to broaden the sixth-form school curriculum. The comment is made that a factor which militates against any broadening of the curriculum is the high standard of specialist knowledge required by the universities for entry to undergraduate courses. While this is in large measure true, it must be realized that there are in fact two related factors. The first is the admittedly high standard of knowledge required and the second is the extreme pressure of applications for the available places in the universities. University places are highly competitive and the better the pass the better the chance of acceptance. It may well be that sixth-form work in schools is bedevilled more by the need to limit objectives, in the hope of achieving a high place in the examination, than by the intrinsic requirements of the examination syllabuses themselves.

Another development in schools which Sir Willis notes with disapprobation is the introduction of engineering subjects into the sixth-form curriculum. These, he suggests, are possibly introduced with the aim of stimulating interest in engineering as a career and providing an incentive to serious study. It would, however, be through a misconception of the educational needs of the professional engineer, if grammar schools were to encourage boys with little aptitude for mathematics and the basic sciences to continue at school up to the age of eighteen to study technical subjects. "The profession of engineering is," he asserts, "no haven for the scientific weakling." That is a dictum which cannot be too often reiterated to every schoolmaster.

Science teaching in schools is, however, determined by the availability of suitably qualified teachers, and the continuing shortage of teachers of science dominates all other considerations relating to the development of pure and applied science. Sir Willis Jackson states the facts with a simplicity which signalizes the urgency of the situation. In 1965 the school population of seventeen-year-olds will reach its peak of twice the 1959 numbers. In 1958, there were, in maintained secondary schools, some 8,900 teachers of mathematics, physics or chemistry, but more than 800 posts in these subjects were either vacant or unsatisfactorily filled. While there is a small but definite improvement in the rate of recruitment to school teaching among science graduates, this will be completely outstripped by the rate of increase of the corresponding school population, altogether apart from the leeway which still requires to be made up. Allowing that there is considerable reserve of capacity resulting from the small numbers in many sixth-form classes, it is none the less evident that the shortage must become acute. Referring to this problem, Sir Willis comments that salary is by no means the only consideration that affects the willingness of science graduates to enter the teaching profession. Teaching facilities, amenities and general conditions of employment are also important factors. It is worth while in this connexion to note that the shortage of teachers in technical colleges is less acute than in schools. Salary scales are rather better, but equally or even more importantly, the technical college teacher has greater freedom from incidental duties, has first-class laboratory facilities and he is encouraged to pursue research. In individual instances pupils have transferred from schools having inadequate science teaching to neighbouring technical colleges where fully satisfactory facilities have been available. While such transfers of pupils on any substantial scale would subvert for the time being the policy of building up the sixth-form science teaching in schools, their utilization as a temporary emergency measure should not be overlooked.

The number of undergraduate students of science and technology in the universities of Great Britain has increased by a factor of nearly three since 1938–39 and now constitutes about 38 per cent of the total university student population of almost 100,000. For the most part these students pursue courses of three years duration. Of these undergraduates, about 40 per cent study engineering or some branch of applied science; but it must be remembered that an appreciable proportion of graduates in pure science, particularly in physics and chemistry, become technologists. The Institution of Electrical Engineers accepts into its membership an increasing number of such physicists.

Sir Willis Jackson takes note of the pressure to increase the length of the undergraduate course, a pressure resulting from the remarkable expansion of the field of knowledge and from recognition of the undesirable degree of specialization existing both in school and undergraduate courses. He does not, however, concede the point that if an extension were practicable it would necessarily be desirable to make it. He considers that for the majority of students of technology three years in the university constitutes a long enough preparation for active participation in the affairs of life outside. In his opinion it is of greater importance to examine afresh the objectives and content of university courses in technology in the light of the new situation, in which a substantial contribution to the teaching of technology at university-level is being made by the colleges of advanced technology. This is indeed a matter of first importance. The universities and the colleges of advanced technology are partners in the education of that section of the youth of Great Britain destined to become the professional engineers and applied scientists.

The new diploma in technology has been conceived and developed with the underlying assumption that there is an approach to the study of applied science which has more emphasis on application than is appropriate to universities, but which can still constitute an academic discipline of university rank. It is believed that by virtue of the integration of studies with industrial experience, the student will be better able to appreciate aspects of technology and to approach, with greater understanding, subjects related to the organization of industry. This is the factor which gives its distinctive quality to the diploma in technology. The validity of this assumption is being put to the proof in courses which are now running in the eight colleges of advanced technology and in the regional colleges which have so far been established. There is encouraging evidence that these courses are justifying the hopes of their designers. At the same time, it is clear that a conscious and sustained effort will be required to ensure that courses for the diploma in technology and courses in universities supplement one another and that, by fostering and emphasizing their distinctive qualities, the two combine to augment the national total of applied scientists. Sir Willis implies that both the universities and the colleges of technology should give serious thought to identifying what are to be the essential distinctive qualities of their courses so that the danger may be avoided of both types of institution offering courses which academically are scarcely distinguishable.

Postgraduate study and research are subjects also very relevant to the making of professional engineers. Research in one form or another is the vital force of academic life, whether the academy be a university or a college of technology. The spirit of inquiry which should characterize the outlook and inspire the teaching of the staffs of universities and colleges requires for its nurture not only a favourable intellectual climate but also the physical resources for its growth. The pattern of the research schools in the universities is essentially in the classical tradition in the sense that the master gathers about him a group of the most able young scholars who, on one hand, draw inspiration from the master and on the other furnish him with the stimulus of their fresh intellects and the challenge of their problems. Research in the physical sciences requires facilities and services of increasing magnitude and complexity, and when the sciences are applied sciences, the situation becomes vastly more complicated by the interrelation between practice and science. Here unquestionably the stimulus of achievement in application can match and even surpass that of intellectual curiosity.

It is, in the opinion of Sir Willis Jackson, quite unrealistic to suppose that the teaching of technology at its highest level can be carried out wholly or largely by men who occupy full-time teaching posts, unless these men return to industry or a similar environment at appropriate intervals. He commends the practice, long established in other countries, by which, for limited periods, carefully selected experts from industry or government establishments are given academic status and contribute to the advanced specialist teaching of technology. Drawing a parallel with the position of medical clinicians, who carry full academic standing in the universities, he goes on to suggest that the separation of establishments of the Department of Scientific and Industrial Research from the domain of technological teaching constitutes a serious national handicap. It would, he considers, be a useful first step if these research establishments and some of the research associations could be accepted as recognized institutions of universities in the sense that selected members of their staffs could be fully recognized as teaching members of appropriate faculties.

That the highest level of knowledge of the specialized applications of science to technology resides in industrial and government research establishments and design departments is undeniable. It is, however, equally true that there is a network of informal association between the staffs of universities and colleges, and those of industrial organizations and government establishments. The extent and importance of this must not be under-estimated. Exchange of information, participation in seminars, membership of research committees and the provision of specialist lecturers—all these feature in an informal and cordial co-operation from which the greatest benefit is derived. It is a widening of this association and its organization on a more formal basis which Sir Willis Jackson advocates. The practice in the technical universities of the Continent of Europe of appointing as professors distinguished engineers and scientists in industry who then divide their time and attention between their academic and their industrial responsibilities possesses the great merit of bringing current industrial problems directly to the notice of students. The Continental system, however, has its drawbacks. Faculties are much less closely knit than in Great Britain, and co-ordination of teaching is, by comparison, relatively slight. Not infrequently academic visitors from the Continent remark with appreciation upon the unity and coherence not only of faculties but also of complete universities in Great Britain. If a university department were to lean unduly upon the support of a group of experts from outside, it would be in danger of weakening that intellectual integrity and coherence which are its priceless possessions. Undoubtedly the strengthening of the link between technological teaching and industry is of major importance. The success of whatever methods are adopted to that end will be gauged by the growing intrinsic strength of university and college departments

It is only relatively recently that universities and colleges have made regular and systematic provision for postgraduate study. Sir Willis stresses that the need for postgraduate courses will grow, and that participation in providing them must be recognized and accepted as an increasing necessity. Referring to postgraduate study in university departments of engineering, he subscribes to the view that a period of industrial training is an essential pre-requisite to the choice of a field of advanced work and to effective study in it. That industry has made only a very limited response to the offers of courses of postgraduate study which have so far been made by universities is, in Sir Willis's opinion, due in part to the shortage of graduates at a time of expanding industrial activity and in part to a measure of inflexibility in universities themselves. Courses of shorter duration than a full university session would, he believes, have been more attractive.

Sir Willis does not, however, refer to a feature of postgraduate study which is of considerable significance, namely, that there are in fact two fairly well differentiated types of postgraduate course which might be described as specialist and fundamental. The specialist course deals with a branch of technology in which advanced methods of analysis and design have been formulated, the significance of which will be best appreciated by someone already familiar with the practice of the subject. The fundamental course, on the other hand, simply represents an extension of the undergraduate course. The potential research engineer who has completed the normal undergraduate course in engineering starts his industrial career with a severe handicap in that he possesses only the most superficial knowledge of modern physics. In all probability he also has but little familiarity with the methods now available for tackling intractable problems with the aid of computers. These are examples of fundamental matters, an appreciation of which is not critically dependent upon industrial experience; equally importantly, they constitute rigorous studies to which the mind is unlikely ever to be better attuned than at the end of the undergraduate course. There are cogent arguments in favour of accepting for such fundamental courses, immediately after graduation, those honours graduates who show real aptitude for theoretical studies.

In the provision of postgraduate courses on a part-time basis, the major technical colleges of Great Britain have a noteworthy record. The new colleges of advanced technology are building up rapidly their activities in the postgraduate field, and their close association with industry places them in a very advantageous position for doing so. Research projects undertaken with the view of qualifying for the new higher award of the National Council for Technological Awards will normally be carried out mainly in industry but in close association with the teachers of one of the colleges of advanced technology. It is a happy augury for the future co-operation of universities and colleges of advanced technology that the postgraduate award of the National Council, the membership of the College of Technologists, will be available to suitably qualified university graduates and, equally, the universities will accept suitable holders of the diploma in technology as postgraduate students pursuing courses of postgraduate study or research leading to higher degrees. There is here an opportunity for forming a most fruitful association between the two types of institution. Just as at the undergraduate-level there will be a broad overlap in the courses themselves and in the qualities and the abilities of the men who pursue them, so in the postgraduate field will there be many courses of study and many research projects which could be pursued equally well in either type of institution. None the less, if the theory of the diploma in technology is well founded, there will be a general bias towards more applied problems in the colleges of technology and towards more fundamental problems in the universities. Common seminars, joint discussions and the occasional sharing of experimental facilities, will disclose a great community of interest and serve to engender that mutual respect which is the basis of true friendship.

The professional engineer requires practical training in addition to his academic qualifications, and Sir Willis Jackson devotes a good deal of attention to this subject. The fact is that the practical training facilities of Britain are already inadequate in scale, and Sir Willis describes the shortage of these facilities as the Achilles heel of our national plans for the further development of technical and technological education. The large manufacturing organizations have well-developed schemes of graduate apprenticeship, but the number of young graduates requiring training is already in excess of the number which these companies can handle satisfactorily. Sir Willis advocates the more active development of group training schemes in which smaller and more specialized firms co-operate in a scheme in which their limited individual resources are combined to provide a balanced training.

Difficulty in obtaining satisfactory practical training is a matter which affects very directly the Dip.Tech.(Eng.), for the qualification certifies not only that its possessor has attained the requisite academic standard but also that he has satisfactorily completed the required period of practical training. The problem of ensuring that this training has been satisfactorily carried out and that a proper balance has been maintained among its constituent parts is not easy of solution. It does, however, demand the closest attention, for a reliable method of assessment of practical training will ensure for the diploma in technology, in its training aspect, the standing and recognition which it merits.

Dealing with the utilization of our scientific man-power, Sir Willis considers that the efficient use of professional engineers and scientists is severely handicapped by failure to tackle effectively the education and training of the craftsmen and technicians by whom they should be supported. In addition to that, there is, he believes, much duplication of effort in research and design departments within the large organizations and between manufacturing industry and the research associations. This dissipation of effort reduces the resources which could be mobilized to launch a powerful attack on a new field.

Shortage of university places and limitations in the availability of practical training facilities may at first sight appear a fairly domestic matter. The fact is, however, that these limitations react very seriously on our ability to provide education and training for potential engineers and scientists from the Commonwealth and particularly from the under-developed countries. As a contribution to international co-operation and as a tangible expression of good will, a substantial increase in our education and training facilities, provided specifically for overseas students of technology, would do much to strengthen our national self-respect and would redound in more senses than one to our credit.

SCIENTIFIC CENTENARIES IN 1960

By JOAN M. EYLES

THE contributions to science made by royal persons lie more in the realm of patronage than in the performance of scientific work. One recalls the association of Charles II with the Royal Society, Louis XIV with the Royal Academy of Sciences in Paris and Frederick I with the Berlin Academy. Nevertheless, there have been some notable exceptions. The two Medici brothers, Grand Duke Ferdinand II and Leopold, were deeply interested in science, had a laboratory, and were responsible for the formation of the Academia del Cimento. Both Prince Rupert, cousin of Charles II, and Philip, Duke of Orleans, brother of Louis XIV, devoted much of their time to scientific study; while in Scotland James IV took part in alchemical operations.

This year, in Lisbon, an International Congress of the History of Discoveries, to be held under the auspices of the Portuguese Government, will commemorate the five hundredth anniversary of the death of another royal scientist, Prince Henry the Navigator. Prince Henry, the third surviving son of King John I of Portugal, was half English, for his mother was a daughter of John of Gaunt, the fourth son of Edward III. Prince Henry devoted his life to organizing voyages of exploration, both along the west coast of Africa and to the different groups of islands in the Atlantic. He resided for more than twenty years at Sagres, close to Cape St. Vincent, whence he could look out over the Atlantic. Here he briefed his captains and studied their reports. He employed skilled mathematicians and astronomers to instruct his navigators and to prepare charts, so that under his supervision Sagres became a centre of geographical research. The proceedings of the Congress should throw new light on this important era of discovery and colonization.

Another outstanding man with a gift for organization, this time an English physician, was Thomas Linacre (1460–1524). His name will always be associated with the foundation, in 1518, of the College of Physicians of London, of which he was the first president, meetings being held at his house. He also endowed three lectureships in medicine, two at Oxford and one at Cambridge. Linacre, who was physician to Henry VIII, and to many other great men of the time, had studied medicine at Padua. His only medical writings were translations of a number of works by Galen; but he was also the author of a Latin syntax and two elementary grammars, and he had a high reputation as a Greek and Latin scholar.

The sixteenth century saw the beginnings of British attempts at colonization of the North American coast. In 1585 a large party was sent by Sir Walter Raleigh to form the first colony. With them went Thomas Hariot, the man of science chosen by Raleigh to make a survey of the new land, and to report on the mineral resources, as well as plant and animal life. Hariot, born in 1560, and already distinguished as a mathematician, made a report, published in 1588, entitled "A Briefe and True Report of the New Found Land of Virginia", which is now a very rare book. Afterwards, he resumed the study of mathematics, and made important contributions to the theory of equations; and he was the first to use the two signs of inequality. His book on algebra, "Artis Analyticae Praxis", was not published until 1631, ten years after his death.

Contemporary with Hariot was the Swiss botanist Caspar (or Gaspard) Bauhin, born at Basle in 1560 His principal work, "Pinax Theatri Botanici", in which he described some six thousand species, appeared in 1623, a year before his death. Bauhin did invaluable work in disentangling the many botanical synonyms in use at that time and also made one of the first attempts at systematic classification. He abandoned the method of describing plants in alphabetical order of name, and used instead a binomial nomenclature, in which he anticipated Linnaeus.

This year is the tercentenary of the death of the mathematician who invented the slide rule, and also introduced those familiar signs, × for multiplication and : : for ratio. This was William Oughtred, born at Eton, where his father was a master in the College. He was elected to a fellowship in King's College, Cambridge, at the age of twenty. He was devoted to mathematical studies, and the time spent on them, he said, "I redeemed night by night from my natural sleep, defrauding my body, and inuring it to watching, cold and labour, while others took their rest". Most of his life was spent as rector of Albury, in Surrey, where many students of mathematics, including Seth Ward, Wallis and Wren, visited him for instruction and discussion. Oughtred was the author of a highly esteemed text-book of arithmetic and algebra, "Clavis Mathematicae", which went into a number of editions, and was later translated into English with a preface by Halley.

Two men, both of whom became professors of medicine at the newly founded University of Halle, and who shared an interest in chemistry, were born in 1660. The senior of the two was Friedrich Hoffmann, who gave his name to Hoffmann's anodyne and carried out much analytical work on mineral waters. He visited England, and was elected a Fellow of the Royal Society in 1720. He had as assistant Georg Ernst Stahl, whose name will always be linked with the phlogiston theory. These two supported opposing schools of philosophy, Hoffmann being a mechanist, and Stahl a vitalist. This led to strained relations between them, and in consequence Stahl resigned his professorship, and became Court physician in Berlin, where he died in 1734. Hoffmann died six years later.

Another physician whose interests were not restricted to medicine was Sir Hans Sloane (1660–1753), president of the Royal Society during 1727–41, whose collection of books, manuscripts and natural history specimens, together with the Harley and Cotton collections, formed the foundation of the British Museum. Sloane lived to be nearly ninety-three, and it is only seven years since the bicentenary of his death was commemorated.

The British Museum was first opened to the public in 1759, but seventy-six years earlier there was opened in Oxford, a museum, the Ashmolean, which contained not only collections open to public view, but also a laboratory and school of natural history. The first keeper of this Museum was Robert Plot, author of the "Natural History of Oxfordshire". The second was his assistant Edward Lhuyd, the tercentenary of whose birth also falls this year. Lhuyd was a Welshman whose interests included philology and archæology, as well as botany and geology. He made a considerable collection of fossils, and prepared an illustrated catalogue of them, which was published in 1699. It was the first systematic catalogue of British fossils. The University had refused to publish it; but ten subscribers, including Hans Sloane and Isaac Newton, put up the money for its publication, and it appeared in a small edition of 120 copies. Lhuyd died at the early age of forty-nine, the victim of a chill caught, it is said, through sleeping in a damp room at the Museum

Among the scientists who died in 1760 four call for particular notice—an astronomer, an anatomist, a botanist and a chemist. The astronomer, Louis Godin, was one of a party sent in 1735 by the Royal Academy of Sciences in Paris to measure the length of a degree near the equator. After some years in Peru, Godin, having completed his mathematical work with the expedition, accepted a post as cosmographer at the University in Lima He died at Cadiz in 1760, aged fifty-six.

The anatomist, J. B. Winslow, was born in Denmark in 1669, but became a naturalized Frenchman, and was also a member of the Royal Academy of Sciences in Paris. His lectures at the Jardin du Roi attracted large audiences, and his text-book of anatomy was in use for a century. He died in France at the age of ninety-one.

The sexual system of plant classification advanced by Linnaeus was not accepted at the time by all botanists, and one of its most persistent opponents was Charles Alston, professor of botany and materia medica at Edinburgh from 1710 until his death in 1760. Alston had studied at Leyden under Boerhaave, and his opinion carried sufficient weight to prevent any general acceptance of the Linnaean system in Scotland until after his death.

In Germany, J. T. Eller (1689–1760), physician to Frederick the Great, was one of the many eighteenth-century medical men who devoted much of their time to the study of chemistry. He carried out work on the solubilities of salts, and issued many papers in the publications of the Berlin Academy. Eller was also an influential supporter of Stahl's phlogiston theory.

Stahl's theory was soon to be opposed by the revolutionary ideas of Lavoisier, and two chemists born in 1760 became teachers of the new chemistry in their own countries. Thomas Beddoes, born in Shropshire, is remembered to-day more for his discovery of the young chemist Humphry Davy, whom he installed as superintendent of his Pneumatic Institute in Bristol, than for his chemical work. He was, however, a skilled chemist, and was reader in chemistry at Oxford during 1788–92. Attention has recently been directed to the efforts he made, while occupying this post, to improve the scientific side of the Bodleian Library (Robinson, E., *Annals of Science*, 11, 137; 1955). Beddoes had attended Black's lectures in Edinburgh, and had met Lavoisier in Paris. His own lectures appear to have been very popular, for he wrote to Black in 1788 that he had "the largest class in chemistry that has ever been seen at Oxford within the memory of man, in any department of knowledge". Three years later he wrote to congratulate Black on his renunciation of the "old Chemical Theory", from which it seems clear that Beddoes himself had already accepted the teachings of Lavoisier.

The first Swedish text-book to support Lavoisier's theories was written by Johan Gadolin, who was born at Åbo, in Finland, on June 5, 1760. Gadolin's early work was concerned with specific heat, but later he devoted himself to chemistry. He studied at Uppsala, and travelled widely in northern Europe. In 1794 he investigated an unusual mineral that had been found some years before at Ytterby, near Stockholm, and separated from it the first of the rare earths, later named yttria. Afterwards the mineral in which it was found was named gadolinite, by Klaproth. In 1797 Gadolin was appointed professor of chemistry at the University of Åbo, a post which he held for twenty-five years. He died in 1852, at the age of ninety-two.

Passing on to those who died only a century ago, we note first the brilliant young Hungarian soldier and mathematician, Janos Bolyai (born 1802). One of the first to challenge the authority of Euclid, he constructed a non-Euclidean geometry, which was published as an appendix to a text-book of geometry by his father, also a mathematician, in 1832. Lack of recognition, and the translation into German of a similar work by Lobachevsky, which had appeared in Russian in 1829–30, disappointed Bolyai, whom Gauss described as a "genius of the first rank", and he published no other mathematical work.

Another professional soldier who also made a name in science was General Sir Thomas Makdougall Brisbane, governor of New South Wales during 1822–26. He established an observatory at Paramatta in 1822, and the Brisbane catalogue of stars published in 1835 was the result of observations made there. Succeeding Sir Walter Scott as president of the Royal Society of Edinburgh in 1833, he occupied the chair until his death in 1860. He was also president of the British Association at its fourth meeting, in Edinburgh, in 1835. Important magnetic observations were made at his observatory at Makerstoun in Scotland, gaining for him the

Keith Prize of the Royal Society of Edinburgh in 1848.

A valuable step towards the theory of evolution was made when M. H. Rathke (born 1793), a native of Danzig, discovered the fish-like gill-slits and their blood vessels in the embryos of birds and mammals. Rathke's extensive writings include many embryological studies, the first detailed anatomical description of *Amphioxus* and accounts of many other marine animals. He succeeded the pioneer embryologist Von Baer at Konigsberg, where he remained until his death in 1860. He was elected a foreign member of the Royal Society in 1855.

Anthropology as a science can be said to have been established by J. F. Blumenbach of Gottingen, and his measurements of skulls entitle him to be regarded as the founder of craniology. It was not, however, until the Swedish biologist Adolf Anders Retzius (1796–1860) invented the cephalic index and used it as a basis for classification that craniology achieved a modern aspect. Retzius was born at Lund, and was one of three generations of distinguished natural scientists. He carried out biological research in many fields, and pioneered the study of comparative anatomy in Sweden.

The centenary of the birth of three well-known physiologists, two British and one Dutch, falls in May 1960. William Einthoven, born in Java, was professor of physiology in the University of Leyden for nearly forty-two years. He had a gift for solving physiological problems by physical methods, and in 1913 he invented the string galvanometer, and applied it to investigating the electric phenomena of the heart-beat. This led to the development of the electrocardiograph. In 1924, three years before his death, he received the Nobel Prize for Medicine. In 1926 he was elected a foreign member of the Royal Society.

Both the British physiologists, W. M. Bayliss and J. S. Haldane, did valuable work during the First World War, Bayliss in the treatment of wound shock, and Haldane, who had studied the respiratory processes under widely varying conditions, in the treatment of gassed patients. Haldane, who became director of the Mining Research Laboratory, was always particularly interested in problems affecting the health of miners. He received the Copley Medal of the Royal Society in 1934, two years before his death. Bayliss, well known as the author of "Principles of General Physiology", made an important contribution to science when, in 1902, with Starling, he announced the discovery of secretin, the first of the hormones to be discovered. He was elected a Fellow of the Royal Society in 1903, received the Copley Medal in 1919, and died in 1924.

Another Nobel prize winner in medicine was N. R. Finsen, the Danish physician, who was born in the Faeroes in 1860. He discovered the curative effects of ultra-violet light, and was the first to use sunlight baths in the treatment of tuberculosis. He died a year after the award was made, when only forty-four.

Turning to chemistry, yet another recipient of a Nobel award was the German chemist Eduard Buchner, who died near Verdun, on the Western Front, during the First World War. In 1897 he expressed enzymes from yeast cells and showed that alcoholic fermentation was caused by chemical agents, and not by physiological processes in yeast cells, as was previously believed. He was awarded the Nobel prize for chemistry in 1907.

An English chemist born in 1860 who cannot be overlooked is W. H. Perkin, junior, brilliant son of a brilliant father, who was professor of chemistry first in the Heriot-Watt College in Edinburgh, thence moving to Manchester, and finally becoming Waynflete professor of chemistry at Oxford in 1913. Here he remained until his death in 1929. Perkin carried out many important organic syntheses, and did much to advance the study of chemistry at Oxford.

To attain outstanding distinction both as a scientist and as a classicist is unusual, but it is one of the claims to fame of Sir D'Arcy Wentworth Thompson, for he was a Darwin medallist of the Royal Society and also served as president of the Classical Association (similarly Sir Archibald Geikie, the geologist, served as president of the Classical Association, and Sir Cyril Hinshelwood is now president both of the Royal Society and of the Classical Association). D'Arcy Thompson owed his deep knowledge of the classics to his father, professor of Greek in Queen's College, Galway. He studied medicine at Edinburgh, his birth-place, and biology at Cambridge, where he took both parts of the Natural Science Tripos with first-class honours. When only twenty-four, he was appointed professor of biology in the newly founded University College of Dundee; and there, and at St. Andrews, he remained actively at work until his death in 1948. Although his most outstanding work is his book "On Growth and Form", his volume of essays entitled "Science and the Classics" is more representative of his wide range of interests.

There must be many old students of D'Arcy Thompson who remember him with affection and gratitude, for he loved teaching, and the same can be said of the geologist W. W. Watts (1860–1947), who became professor of geology in the Imperial College of Science and Technology (then the Royal College of Science) in 1906, and over a period of twenty-four years built up the present well-known school of geology. Although his devotion to teaching left him little time for research, he carried out valuable field-work in Shropshire and in Charnwood Forest. "Geology for Beginners", the little book he first published in 1898, is still in print, and has probably been used by more elementary students of geology than any other work. Watts was president of the British Association in 1935, and in 1945, at the age of eighty-five, he delivered the first William Smith Lecture to the Geological Society of London.

Another worker in the field of geology and geography was Sir Sidney Burrard, who joined the Survey of India in 1884, and in 1901 published a paper on "The Attraction of the Himalayan Mountains on the Plumb Line in India", which threw new light on the distribution of gravity anomalies. Burrard was surveyor-general of India during 1911–19 In 1913 he received the Victoria Medal of the Royal Geographical Society for his work in India.

Other scientists born in 1860 include E. A. Sperry, American inventor of the gyro-compass; D. E. Smith, historian of mathematics and founder in 1924 of the History of Science Society in Boston; W. B. Spencer, the anthropologist; and J. G. Bartholomew, the Scottish cartographer and one of the founders of the Royal Scottish Geographical Society.

No reminder should be needed that 1960 is the tercentenary of the first formal meeting, on November 28, 1660, of the Royal Society, although the Royal Charter was not obtained until 1662.

OBSERVATIONS RELATED TO THE IMPACT OF *LUNIK II*

By G. FIELDER

Director, Lunar Section, British Astronomical Association

THE accompanying table gives particulars of observations which may be related to the impact of *Lunik II* with the Moon.

These observations refer to at least four different spots on the Moon's surface and cannot all refer to the impact of the instrument container of *Lunik II*. However, more than one object may have hit the Moon. From the point of view of position, it will be noted that most of the observations are consistent with the radio result. Those not consistent in this respect are asterisked. Photographs of moderate quality taken from the Pic-du-Midi, and from G. A. Hole's Observatory at Patcham, before and after the impact of the instrument container, do not appear to confirm any of the above observations.

OBSERVATIONS WHICH MAY BE RELATED TO THE IMPACT OF *Lunik II*

Observer(s)	Location	Telescope	September 13, 1959 Univ time	Position on Moon	Description of phenomenon (and its duration)
J. G. Davies A. C. B. Lovell (ref. 1)	Jodrell Bank	250-ft. Radio-telescope	21h 02m 23s.	Within 7′ arc from centre of Moon's disk	Velocity of impact, ~ 3 ± 0·5 km./sec. (instantaneous cessation of radio signals)
P. Moore (ref. 2)	E. Grinstead	12½-in Reflector, × 300–400	21h. 02m 23 ± 2s.	+ 085, + 195	Pinpoint of light (< 0·5 sec.)
H. P. Wilkins (ref. 3)	Bexleyheath	15½-in. Reflector, × 300	21h 02m 23s.	Close to Schneckenberg	Pinpoint of light and dark ring (few seconds)
B. Balázs J. Balázs M. Lovas	Konkoly	7-in. Refractor	21h. 02m. 30s.	+ 024, + 434	Dark spot, 4 km. in diameter, expanding asymmetrically into a hazy, grey object elongated to N. Mean width then ~ 40 km. (14 min.)
P. A. Murray	Carrickfergus	6-in. Reflector, × 112	21h. 02m. 15s.	Auwers	Distinct brightening (1·5–2 sec.)
R. D. Williams	Carrickfergus	Small refractor, × 30	21h. 02m. 15s	~ Auwers	
S. Bradford*	South Shields	3-in. Refractor, × 120	22h. 03m.	Littrow	Dusky shading (> 17 min.)
A. Florsch	Strasbourg	6½-in Refractor, × 150	Some time between 21h. 01m. and 21h 05m	+ 340, + 250	Patch becoming as large as Plinius *A* and doubling its diameter in 3–4 min of time
R. G. Townsend*	Sanderstead	1 in Refractor, × 15	21h. 02m	Near junction of M. Seren. and M. Tran.	Black spot appearing to diffuse rapidly (~ 5 min.)
R. P. Townsend*	Sanderstead	2-in. Refractor, × 20	21h. 02m.	In vicinity of Littrow, Mac-Robius, and Römer	Jet black spot, expanding and turning dark grey (> 5 min.)

[1–3] *Nature*, **184**, 501 (1959)

NEWS and VIEWS

National Institute for Research in Dairying: Dr. A. T. R. Mattick

DR. A. T. R. MATTICK, until September 30 deputy chief scientific officer on the staff of the National Institute for Research in Dairying at Shinfield, retired on that date, having reached the age limit. Apart from the period 1915–19, when he served in the First World War in the Royal Army Medical Corps, the whole of his working life has been spent at the Institute, which he has seen grow from very small beginnings to its present status as the outstanding dairy research institute in the Commonwealth, if not in the world. In 1919, he became a graduate assistant to Dr. R. Stenhouse Williams, and succeeded the latter as head of the young Institute's Bacteriology Department in 1929. He became deputy director in 1948, still retaining the headship of the Bacteriology Department. In October 1958, on the retirement of Prof. H. D. Kay, he became acting director.

The development, during the past thirty years, of sound methods of dairy hygiene, both in Britain and in countries abroad owes a very great deal to Mattick and his colleagues. When Mattick took it over, his Department at Shinfield had a scientific staff of three only, and its resources were meagre. With increasing funds and accommodation since 1933, and a carefully selected, growing staff, the early pioneer work was extended not only to cover the wide field of the application of microbiological science to the main problems of the dairy industry, but also to make substantial contributions to our basic knowledge of micro-organisms. Thus, on one hand, his laboratory was largely responsible for defining the exact conditions of heat exposure necessary in the continuous process now almost universally employed in Britain for the pasteurization of milk (conditions duly incorporated into the official regulations), and on the other hand, for the detection, isolation and identification of the interesting and powerful antibiotic nisin, produced by a streptococcus that he isolated from milk, and now being used industrially. These are but two examples from many that could be mentioned of the types of problems tackled under his direction. Most of the leading dairy bacteriologists in Britain, and many of those in other countries, have engaged in research for longer or shorter periods in his laboratory. His training there of these young and not-so-young postgraduates laid the essential foundation for the advisory bacteriological branch of the National Agricultural Advisory Service. Mattick retires from a strong, active department, now of fourteen graduates and twenty-four assistants, not only as the acknowledged doyen of British dairy bacteriologists but also with a world-wide reputation as an outstanding research worker in pure and applied microbiology. His able successor as head of the Bacteriology Department at Shinfield is W. A. Cuthbert, until recently a senior bacteriologist in the National Agricultural Advisory Service.

Preventive Medicine at Sheffield:
Prof. John Knowelden

DR. JOHN KNOWELDEN has been appointed to the chair of preventive medicine and public health in the University of Sheffield. Educated at King's College, London, and St. George's Hospital Medical School, London, Dr. Knowelden qualified in 1942, and after a short period as house surgeon at St. George's Hospital, he was a surgeon lieutenant in the R.N.V.R. during 1942–46. In 1947 he graduated M.B., B.S., in the University of London and was also awarded the D.P.H. with distinction. In the same year he was awarded a Rockefeller fellowship in preventive medicine, which he held during 1948–49 at the Johns Hopkins School of Hygiene and Public Health, Baltimore. In 1949 Dr. Knowelden was appointed to his present post of lecturer in the Department of Medical Statistics and Epidemiology at the London School of Hygiene and Tropical Medicine. Dr. Knowelden is a member of the Medical Research Council Statistical Research Unit and has been secretary of a number of committees of the Medical Research Council, including the Poliomyelitis Vaccines Committee which planned and executed the 1956 field-trials of British poliomyelitis vaccine. Dr. Knowelden is a joint editor of the *British Journal of Preventive Medicine*.

American Geographical Society: Awards

THE following awards have been announced by the American Geographical Society: *David Livingstone Centenary Medal*, awarded by the Society "for scientific achievement in the field of geography of the southern hemisphere", to William E. Rudolph, of New York City, former chief engineer of the Chile Exploration Company and now managing consultant of Dorr Consultants; *Cullum Geographical Medal* to Dr. Albert P. Crary, chief scientist for the U.S. Antarctic Research Project, for geophysical research in both the North and South Pole regions, and for geographical discoveries; *Daly Medal* to Dr. Richard Hartshorne, professor of geography at the University of Wisconsin, for his contributions to political geography and to "the history and methodology of geographic thought".

University Grants Committee: Appointments

To fill vacancies on the University Grants Committee, the Chancellor of the Exchequer has made four new appointments. The new members, whose appointments will expire on December 31, 1964, are: Sir Eric Ashby, master of Clare College, University of Cambridge; Prof. F. W. Rogers Brambell, Lloyd Roberts professor of zoology, University College of North Wales, Bangor; Prof. C. H. Philips, professor of oriental history and director of the School of Oriental and African Studies, University of London; and Prof. J. Monteath Robertson, Gardiner professor of chemistry, University of Glasgow. The retiring members of the Committee are: Prof. R. H. Matthew, professor of architecture, University of Edinburgh; Prof. P. B. Medawar, Jodrell professor of zoology and comparative anatomy, University College, University of London; Prof. A. Robertson, previously Heath Harrison professor of organic chemistry, University of Liverpool; and Sir George Thomson, master of Corpus Christi College, University of Cambridge, and emeritus professor of physics, University of London.

Zenith Reactor commences Operation

THE *Zenith* reactor at the Atomic Energy Establishment, Winfrith, Dorset, started operating for the first time on December 19. The final stages of commissioning the plant will be carried out during the next few months. The reactor has been built to operate under temperature conditions similar to those expected in a high-temperature gas-cooled power reactor. The reactor core consists of a nearly cylindrical assembly of 235 fuel elements, 8 ft. high and 4 ft. in diameter, in the form of graphite cylinders, containing pellets of fissile uranium oxide, fertile thorium oxide and graphite. The ends of the fuel elements are made of graphite which, with a 3-ft.-thick graphite annulus surrounding the fuel, form a reflector to return some of the neutrons escaping from the fuel into the core, so reducing the quantity of fuel used. The reactor core is enclosed in a pressure vessel through which nitrogen is pumped in a continuous closed circuit. The nitrogen is heated by a 200 kW. heater as it enters the core: the hot gas raises the mean temperature of the core to 800° C. At the top of the core the hot nitrogen is mixed with cooler gas before passing through the reflector: thus the reflector is kept at a mean temperature of 400° C. The reactor pressure vessel and associated plant are enclosed in an airtight containing building. The reactor itself will be operated at a peak neutron flux of 10^8 neutrons/cm.^{2}/sec. at a maximum nuclear power of 100 W.: the nuclear heat produced is thus negligible in comparison with the energy used to heat the core.

Zenith is controlled by neutron-absorbing rods in the graphite reflector. There are 30 control rod holes disposed in a ring around the core: 20 of these are occupied by motorized control rods and fixed absorbers may be loaded into the other 10 holes. Six flux-measuring chambers are arranged in other holes at the inner edge of the reflector. There are four pulse-counting fission chambers, two for use at high power and two at low power, and also two current fission chambers feeding shut-down amplifiers. Activity in the fuel elements will be at a low level because the reactor power will not exceed 100 W.; this permits easy rearrangement of the core. The reactor cost approximately £400,000, including design, construction and commissioning. For the first two years after commissioning *Zenith* will be used to check reactor physics design calculations for the *Dragon* high-temperature gas-cooled reactor experiment (see *Nature*, 183, 507; 1959).

Upjohn Cell Model at Royal Scottish Museum

FOLLOWING the recent interest in the two B.B.C. television programmes on "Science International", which featured Prof. Michael Swann, of the Natural History Department of the University of Edinburgh, and a group of scientists from world-wide sources, the notable background exhibit, the famous Upjohn model of a living cell magnified by 1,000,000 (Fig. 1), has been transported to the Royal Scottish Museum, Edinburgh, where it will be shown for six weeks. It was specially flown over from the United States for the two 1 hr. broadcasts, and has not been shown to the public anywhere outside the United States until its exhibition in Edinburgh at the middle of December. The model reproduces in 'Perspex', in three dimensions, the accumulated researches into cell structures employing light and electron microscopes over many years. The construction of the model was undertaken by the Upjohn Company, Kalamazoo, Michigan, and the research for it occupied some eighteen months, while the actual building, by Bill Burtin and his team of scientific model designers, took more than eight months. It is a hemisphere of 24 ft. diameter. First shown in June 1958 in San Francisco at the Convention of the American Medical Association, it was moved to a Health Exhibition in the Coliseum, New York City, and then to the Museum of Science and Industry, Chicago, Michigan In the Royal Scottish Museum it is attracting interested crowds ranging from school classes to university —particularly medical—students. In February it is to be flown back to the United States.

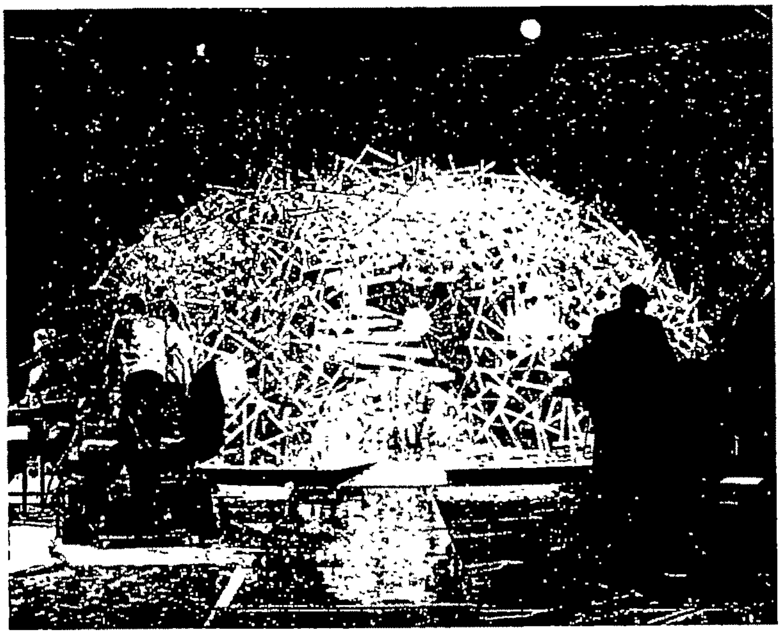
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Fig. 1. The Upjohn cell model

Training in the Use of Isotopes

THE second of the two self-propelled mobile radioisotope laboratories given to the International Atomic Energy Agency by the Government of the United States of America in April 1958 was handed over to an agency representative at the Oak Ridge Institute of Nuclear Studies on December 11. The mobile laboratories are specifically designed to provide the tools for basic training in radioisotope handling techniques. They can tour a geographical area visiting high-schools and universities, and thereby make a training course easily available. At the request of the Government of Mexico, this laboratory will be used at several training establishments and universities in that country during January, February and March 1960. In April, the laboratory will be sent to Argentina for a period of six months.

In Buenos Aires a course has been organized by the National Atomic Energy Commission of Argentina for the training of twenty-five experts from Latin American countries in the use of radioisotopes, especially in medicine and agriculture. For this purpose, the International Atomic Energy Agency is providing the services of two lecturers: Dr. Norman Veall from the Radioisotope Laboratory, Guy's Hospital Medical School, London, and Dr. Robert Harold Wassermann from the Laboratory of Radiation–Biology, Cornell University. Japan has put at the Agency's disposal twenty fellowships of six months duration at the Isotope School of the Atomic Energy Research Institute of Japan, for training in the handling and application of radioisotopes in different fields. The Agency has secured the services of Prof. Milorad Mladjenovic from the Boris Kidric Institute of Nuclear Science, Belgrade, and Prof. James Francis Duncan from the Radiochemistry Department, University of Melbourne.

The first mobile radioisotope laboratory has already been used for training purposes at the University of Vienna, and in Greece. It has just returned to Vienna after having been used at the Haus der Technik, Essen, Federal Republic of Germany, for training young radioisotope specialists, and it is expected to be used in the Far East during 1960.

Physiological Examination of a Live Whale

A 50-TON male finwhale beached on December 5 at Provincetown on the tip of Cape Cod, and Dr. John W. Kanwisher, of the Woods Hole Oceanographic Institution, and Dr. Alfred W. Senft, of the Marine Biological Laboratory, were able to carry out temperature measurements and to obtain a cardiogram before the whale died on the following day. The cardiograms showed a pulse beat of 25 a min. (human, 72 a min.) and an estimated size of the heart of about 225 kgm., compared to some 250 gm. for a human heart. Dr. Senft also determined the time-interval from the pacemaker to the contraction of the ventricle. As the condition of the whale deteriorated, there were changes in the cardiogram not unlike those seen in the human cardiogram when oxygenation is impaired; it appeared that the whale suffered a conduction block, a common defect in man's heart. While the cardiograms were being made, Dr. Kanwisher obtained temperature measurements and collected respiration samples from the whale's blowhole. The internal body-temperature was 92° F., while the fins and tails had a temperature of 50° F.;

initially the dorsal fin (used as a radiator) was much warmer. Gas samples taken from the whale's breath showed that the animal's lungs were not functioning well. It extracted only a third as much oxygen from the inhaled air as a human being does.

National Academy of Sciences—National Research Council

DR. KINZEL, vice-president for research of the Union Carbide Corporation, has been appointed chairman of the Division of Engineering and Industrial Research, National Academy of Sciences—National Research Council, in succession to Vice-Admiral Edward Lull Cochrane. The Division of Engineering and Industrial Research is one of eight divisions of the Academy—Research Council, a private organization of distinguished scientists devoted to the furtherance of science for the general welfare and to advising the Federal Government, upon request, on any matter of scientific or technical interest.

Textile Institute: Officers

DR. A. R. URQUHART, assistant director at the British Cotton Industry Research Association, and honorary secretary of the Textile Institute since 1958, has been elected chairman of the Council of the Textile Institute, in succession to Mr. G. Loasby, who has resigned due to business commitments. Mr. J. C. H. Hurd, head of the School of Textiles of Leicester College of Technology and Commerce, follows Dr. Urquhart as honorary secretary of the Institute. Mr. A. Draper, director of William Birtwistle Allied Mills, Ltd., Preston, continues in office as vice-chairman of Council, and Mr. A. F. W. Coulson, director of R. Greg and Co., Ltd., Stockport, continues in office as honorary treasurer.

Harrison Memorial Prize

APPLICATIONS for the Harrison Memorial Prize for research in chemistry are invited, and should be submitted to the selection committee not later than September 1. The prize is open to natural-born British subjects less than thirty years of age on December 1, 1959, and consists of a bronze plaque and a payment of 100 guineas. The selection committee consists of the Presidents of the Chemical Society, Royal Institute of Chemistry, Society of Chemical Industry and Pharmaceutical Society. Further information can be obtained from the President, Chemical Society, Burlington House, Piccadilly, London, W.1.

University News: Belfast

THE following appointments to lectureships are announced: Dr. A. K. Galwey and Dr. B. D. Flockhart (physical chemistry); Dr. A. Scott (zoology); R. Tavener-Smith (geology); and W. E. Glover (physiology).

London

PROF. K. G. DENBIGH, professor of chemical technology in the University of Edinburgh (see *Nature*, 176, 583; 1955), has been appointed to the University chair of chemical engineering science tenable at the Imperial College of Science and Technology from October 1960 in succession to Prof. P. V. Danckwerts, who has taken over the Shell chair of chemical engineering in the University of Cambridge. Dr. G. H. Williams, lecturer at King's College, has been appointed to the University readership in organic chemistry tenable at Birkbeck College.

Announcements

THE Liverpool Geological Society Medal has been presented to Dr. J. C. Harper, senior lecturer in the Department of Geology, University of Liverpool, in recognition of his work on the Ordovician and Silurian faunas of North Wales and Ireland, and his services as an officer of the Society.

MR. J. H. GLOVER has been co-opted as secretary of the Polarographic Society in succession to Mr. W. J. Parker, who has resigned. All communications to the Society including subscriptions should be sent direct to Mr. Glover at 75 Craven Gardens, Wimbledon, London, S.W.19.

THE Institute of Welding has organized a Christmas Lecture with demonstrations, for young people, on "The Magic Arc", which is to be given by Prof. H. O'Neill. The lecture will take place on January 5 at the Institute, 54 Princes Gate, Exhibition Road, South Kensington, S.W.7. Further information can be obtained from the Secretary of the Institute at the above address.

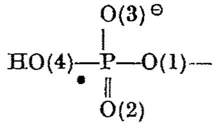
THE Society of Motion Picture and Television Engineers is organizing the fifth International Congress on High-Speed Photography, to be held in Washington, D.C., during October 16–22. In order to facilitate the organization of the Congress, it is desirable that intending authors should present their papers through their national delegate. Summaries must be received by February 16. Further information can be obtained from Dr. R. F. Saxe (United Kingdom national delegate), Queen Mary College, Mile End Road, London, E.1.

A COLLOQUIUM entitled "Proteins of the Biological Fluids" has been organized by St. Jans Hospital, Bruges, during May 6–7. This is the eighth colloquium of its kind to be held by the Hospital. Further information can be obtained from Dr. Hub Peeters, St. Jans Hospital, Bruges, Belgium.

AN International Symposium on Fermentation will be held in Rome during May 9–14, under the auspices of the Istituto Superiore di Sanità (Italian Institute of Public Health), the American Chemical Society (Fermentation Subdivision) and the Società Chimica Italiana. It will consist of scientific papers and panel discussions by invited participants, and is open to all interested scientists. Subjects covered will include fermentation technology (oxygen transfer, fermentor design, etc.), fermentation chemistry and fermentation biology. All inquiries should be addressed to: Fermentation Symposium, Istituto Superiore di Sanità, Viale Regina Elena 299, Rome.

A SYMPOSIUM on 'Inhaled Particles and Vapours' has been organized by the British Occupational Hygiene Society, and will be held in Oxford during March 29–April 1. Further information can be obtained from Dr. J. S. McLintock, Medical Service, National Coal Board, Hobart House, London, S.W.1.

ERRATA. With reference to the communication entitled "Crystal Structure of L-Serine Phosphate" in *Nature* of December 12, p. 1863, Prof. J. Monteath Robertson points out that in the last line on p. 1863, 103 per cent should read 10·3 per cent. Formula II is incorrect and should be:



$$\begin{array}{c} \mathrm{O(3)}^{\ominus} \\ | \\ \mathrm{HO(4)—P—O(1)—} \\ \| \\ \mathrm{O(2)} \end{array}$$

FLORA EUROPAEA IN VIENNA

THE first full meeting of the Flora Europaea Organization was held in Vienna during April 1–7, 1959, at the Naturhistorisches Museum. The international gathering included botanists from Austria, Belgium, Denmark, Finland, France, Germany, Hungary, Iceland, Italy, Jugoslavia, Norway, Poland, Roumania, Sweden, Switzerland, Spain, as well as members of the British Editorial Committee. As editors, advisers and consultants of Flora Europaea they had come to discuss ways and means of consolidating the progress already made in the project and to debate the various theoretical and practical problems which have arisen so far.

The meeting, officially a 'round table symposium', was organized by the Editorial Committee, and the local arrangements were made with the assistance of a preparatory committee of Austrian botanists under the chairmanship of Dr. K. H. Rechinger, director of the Botanical Department of the Museum. The Austrian Ministry of Education contributed to the local expenses, and the International Union of Biological Sciences made a grant to assist members attending. Introductory addresses were given by Prof. H. Strouhal, director of the Museum, Dr. K. H. Rechinger, and the chairman of the Editorial Committee, Prof. T. G. Tutin (Leicester). Prof. Tutin recounted the history of the project, which originated as a result of discussions at the eighth International Botanical Congress held in Paris in 1954. He outlined the principles which had guided the Editorial Committee in its decisions on the size, limits and scope of the work. It was essentially an attempt to summate in one work the essence of taxonomic research made on European plants during the past two centuries, and provide a concise and modern Flora of Europe.

The main work of the meeting was divided into formal sessions in the mornings, during which papers were presented and discussed, and informal afternoon sessions, during which various practical problems were considered. These informal discussions contained much valuable material, which is being edited for inclusion with the main papers in the publication of the proceedings.

The first major topic dealt with was the taxonomy of apomicts. Prof. D. H. Valentine (Durham) gave a paper outlining ways in which the Committee suggested the different kinds of apomictic groups could be treated. It was difficult to lay down general principles for the treatment of all groups since our knowledge of them is so uneven. In dealing with *Hieracium*, *Taraxacum*, *Rubus*, *Ranunculus auricomus*, *Potentilla*, *Alchemilla* and Gramineae, he attempted to set the taxonomic problems against the background of our knowledge of the apomictic mechanisms and the evolutionary potentialities of the groups. Such an evolutionary approach, although not always of direct value, often helped the taxonomist to assess what could be attempted and how far he could go. In *Rubus*, for example, new populations are being constantly produced by hybridization, whereas in most of the *Alchemilla vulgaris* group no hybridization is known. Both groups have been fairly thoroughly investigated by taxonomists, but these evolutionary facts should have a bearing on future attempts to produce workable taxonomic treatments. The problem for Flora Europaea was largely one of devising methods for curtailing and limiting the accounts. Only widely distributed taxa should perhaps be described and endemic taxa dealt with briefly, if at all. All apomictic groups will be dealt with by normal taxonomic methods and will be placed in aggregate species, subspecies or, in exceptional cases, varieties. Experimental categories such as 'agamospecies' will not be used in the Flora, but reference to reproductive mechanisms and other relevant points may be made in the form of observations in the text. This paper was extensively discussed, and particular attention was directed to the procedure of Marklund in his treatment of Fennoscandian *Ranunculus auricomus* representatives, which he grouped into four aggregates which were differentiated morphologically, ecologically and geographically. It was the opinion of several speakers that a similar treatment could be applied to the whole of Europe.

Prof. B. Pawlowski (Cracow) gave the second paper. He made four principal points: (*a*) for apomicts current taxonomic categories should be employed; (*b*) the degree of differentiation of apomicts varies from group to group; (*c*) the most appropriate method of treatment should be left to the author of a particular group to decide; and (*d*) extensive descriptions should be permitted as a departure from the normal abridged ones of Flora Europaea.

In the general discussion the point was made that the level of treatment possible in any apomictic group depended on a number of factors such as the kind of apomixis, the number of taxa likely to be involved, their stability, the treatments already published and how far they could be applied to Europe as a whole. Prof. R. Nordhagen (Oslo) suggested that the term vegetative apomixis was misleading and should be replaced. He believed that phylogenetic lines should be followed in the treatment of such groups as *Taraxacum* and *Hieracium*, but about *Rubus* he would not speak. Prof. H. Merxmuller (Munich) proposed that in the *Ranunculus auricomus* group not only the extreme forms (*R. auricomus* and *R. cassubicus*) should be given, but also 'central' forms under which taxa not belonging to the corner forms could be accommodated. For *Taraxacum* he proposed as models the treatment given by Bocher, Holmen and Jakobsen in "Grønlands Flora" and van Soest's survey of the phytogeography of the genus. The sections recognized are more or less equivalent to aggregates. Keys to this level should be sufficient with further information on apomicts of special interest. For *Hieracium* a possible method would be to follow Janchen in regarding Zahn's *Arten* as aggregates and joining the *Unterarten* in *greges*, the keys not going farther than this level. The number of Zahn's *Zwischenarten* would have to be greatly reduced by the exclusion of primary hybrids. Prof. W. Rothmaler (Greifswald) stressed that apomixis is only one of numerous methods in the plant kingdom and that there was not in principle a basic difference between a persistent inbreeder and an apomict. Normal taxonomic methods should be applied to these groups. Dr. A. Love (Montreal) considered it unfortunate that many taxonomists adhered to the dictum of Winge that the description of hundreds of so-called apomictic species was laying "too much stress on immaterial bagatelles", rather like the collecting of postage stamps. Recent biosystematic studies had shown that apomictic taxa may be of considerable geographical and evolutionary importance. A practical solution could be achieved by combining the system of Turesson (using special categories agamospecies, agamotype, apomict) and that of Marklund. Thus, if the term agamospecies is accepted for apomictic microspecies as approximately the same level as the subspecies proposed by Marklund (and at a lower level than proposed by Turesson) it could be incorporated into the taxonomic system as a category at the subspecific level. The inclusion of only agamospecies at subspecific level would automatically indicate that the species itself is agamospermous, while the inclusion of one or more subspecies also would show that sexual seed formation is also known to occur in the species. Dr. N. Hylander (Uppsala) felt that such a system would not be workable since it was not known in many cases to what extent a species was agamospermous, and therefore such terms as hemiagamospecies would have to be introduced.

The second set of papers dealt with problems of geographical distribution and taxonomy. Prof. H. Meusel (Halle) read a paper outlining his system for indicating the distribution of species by means of formulæ. This system is based on three main elements —countries, floral elements and geographical coordination (floral zones, oceanity-continentality, and 'étages'), and together with distribution maps summarizes a great deal of information in a short space (see Meusel, H., "Arealformen und Florenelemente als Grundlagen einer vergleichenden Phytochorologie" in *Forsch. und Fortschur.*, 33, 163 ; 1959). Meusel's scheme was greatly admired, and it was hoped that it could be extended to the whole of Europe, but it was felt that the limiting factor was the technical one of amassing and processing the data for thousands of species, especially from understudied floras, in time for inclusion in Flora Europaea. Moreover, the number of symbols involved was a serious drawback in a general Flora.

This provoked an illuminating debate about the source and reliability of data used in indicating extra-territorial distributions in Floras in general. The indication in a more or less summarized form of the total distributions of species is almost traditional in most Floras. Dr. V. H. Heywood (Liverpool, secretary of the Editorial Committee) doubted the validity of these data in most cases, since they were almost invariably copied from the literature and errors were thereby perpetuated. Flora Europaea was not intended to be a handbook of plant geography. Prof. D. H. Valentine emphasized the difficulty of knowing whether the species were the same in the various countries from which they are recorded according to these total distributions. Dr. Heywood then talked on some of the problems of taxonomy and distribution in the West Mediterranean, especially the Iberian Peninsula, many parts of which are inadequately known from a floral point of view. This contrasted with taxonomically well-studied areas such as Scandinavia and Central Europe and created grave problems for the authors and editors of the Flora. One result of inadequate exploration in the Iberian Peninsula was that patterns of distribution were far from clear. Gaps may be due to incomplete knowledge and not to the fact that the species concerned show discontinuous areas. The concern here was with basic taxonomy, and there is virtually no cytological or experimental information available. The treatment of three kinds of endemics was considered: endemic vicariant species such as *Ptilotrichum reverchonii* (S.E. Spain) and *P. pyrenaicum* (Pyrenees), where the temptation to unite them should be resisted despite their close morphological similarity; Wettsteinian subspecies showing good geographical pattern with areas of overlap; and micro-endemic vicariants. The latter kind has proved difficult to deal with satisfactorily and is typical of the Mediterranean mountain flora, where each range or peak may have its own endemic representative of a species. Populations have been fragmented into discrete units, but each one is only slightly different from the next, however constant the differences may be. Currently these units are regarded as varieties, subspecies or species, and some uniformity should be introduced to deal with this frequent phenomenon.

The species versus subspecies question was also considered by Dr. K. H. Rechinger in his paper on distributional problems in South-east Europe, where he considered dealing with 8,000 species in the Balkan Peninsula. Although there were many similarities between South-east and South-west Europe, there were a number of differences in climate, physiography, coastlines, etc., which altered the kind of problems found. In the east, one concerned the problems of the island floras with their endemic races, and many years experience had convinced him of the need for a fairly narrow species concept in polymorphic complexes until a detailed revision was possible. Prof. H. Merxmüller (Munich) spoke on the taxonomic treatment of Alpine vicariants, where again the species versus subspecies problem arose. Not all Alpine vicariants could be placed in either category but should be considered for such treatment whenever they showed satisfactory morphological separation coupled with geographical distribution. The important point was to be consistent in their treatment within a group. Too close an adherence, however, to the Wettstein geographical-subspecies method could lead to absurd results whereby species were divided into a series of boxes of little meaning.

The taxonomic treatment of polyploidy was the subject of another set of papers. Prof. W. Rothmaler in his discourse on "Taxonomy and Genetics" said that the working methods and principles of taxonomy must not be mixed with those of genetics. Each taxon corresponds to a concept the extent and importance of which is determined by taxonomic methods. In a species concept, all information, morphological, geographical, ecological and physiological, must be combined with genetical, anatomical and other evidence, but no one line of evidence should be given a greater level of importance than any other. Thus, as regards polyploidy, the chromosome status should be regarded as a valuable aid for the definition of a taxon but should not be given greater importance than any character of internal or external morphology. Similar considerations applied to apomicts. Chromosome numbers may vary as much within a species or subspecies, as they remain constant within a large genus or even part of a family. Dr. V. H. Heywood then gave a paper on "The Taxonomy of Polyploids in Flora Europaea", which was an attempt to suggest practical solutions for dealing with this kind of problem in the Flora. He agreed with Rothmaler's view that no one character or line of evidence is *per se* sufficient to indicate specific status. The bearing of polyploidy in an experimental or genetic special classification is quite different, he suggested, from its value in a taxonomic framework. He dealt with autoploid, alloploid and polyploid complexes, although agreeing that it was not always meaningful or even possible to distinguish between these situations. He proposed that 'cryptic' polyploids should receive no taxonomic recognition, although their existence, and if possible their distribution, should be noted in observations. 'Semi-cryptic' polyploids may be amenable to treatment as varieties, subspecies or in exceptional cases species. Distinct autoploids are undoubtedly rare and should receive specific rank only when they are constantly recognizable; otherwise, if intermediates are known and especially if their geographical areas overlap, they should be given subspecific or varietal rank. Taxa which have arisen by polyphyletic (polytopic) polyploidization should be considered grades *sensu* Huxley and treated as varieties or even forms. Dr. Hylander agreed almost entirely with these views, whereas Dr. A. Löve disagreed almost entirely and proceeded to elaborate his arguments in a paper, "Taxonomy and Chromosomes—a Reiteration". He considered that Flora Europaea should be the beginning of a new era in European taxonomy, not the end of an old outdated epoch. The chromosomes are not just another character comparable to the superficial morphological characters which taxonomists are forced to use in the identification of herbarium material. They determine the characters, whereas the characters do not determine the chromosomes. Changes in the chromosomes determine the divergent evolution of ecotypes and races and then the effective isolation of species, genera and families. The only logical rule for the classification of taxa differing in chromosome number is to name them as distinct species. The ensuing discussion was lively. It was agreed that as much cytotaxonomic information as possible should be given in the Flora, although people were not entirely agreed on its taxonomic importance. A liberal use of species aggregates was a possible solution for morphologically similar (though not identical) cytotypes.

A botanical excursion was arranged by Dr. F. Ehrendorfer on the Monday to Burgenland and Neusiedlersee. The view of the Danube, the large lake and the fascinating salt steppes, combined with the rich spring flora to make it a memorable day. In the evening the participants were given a banquet by the local Government Council at Rust, at which Government Councillor Kunnert mentioned the cultural and scientific traditions of Burgenland—for example, here lived Haydn and the celebrated botanists Clusius and Kitaibel.

The final sessions dealt with the problems of cultivated, escaped and adventive plants. Dr. N. Hylander spoke on his experience with similar problems in the Scandinavian flora. Not only plants of natural habitats but also those growing in localities created or strongly influenced by man had to be considered. Similarly, some of the species growing wild were intentionally or unintentionally introduced by man. It was often impossible to distinguish clearly between these different types. Prof. J. Jalas (Helsinki) gave a paper on "Treatment of the Aliens in Flora Europaea". The alien element could be considered as belonging to the following groups: (1) naturalized or established aliens of extra-European origin; (2) European species occurring in some parts of the area merely as (*a*) naturalized or established aliens, or (*b*) casual adventives. If thoroughly naturalized in one or more European countries the aliens should be described and keyed normally, irrespective of their degree of commonness. Taxa native only outside Europe should be marked with a special sign. The same sign should be used to indicate territories in which native European species are only naturalized. The territories within which a species, whether European or not, occurs as a casual should be enclosed in square brackets. In a second paper he illustrated the valuable data that can be obtained about variable species such as *Medicago lupulina* from countries near the outer limits of their permanent occurrence.

Mention must be made in conclusion of the generous hospitality of the Minister of Education and the Bürgermeister of Vienna, both of whom gave receptions for the participants. A meeting in Vienna would be scarcely complete without a visit to the Wienerwald, and this was also arranged by the city authorities, ending with a Heuriger at Heiligenstadt where, in true botanical tradition, the 'new' wine was liberally sampled. Altogether, the symposium was one of the most successful and certainly the most enjoyable which even Prof. Lüdi could remember.

V. H. HEYWOOD

EFFECT OF SOAP ON THE DIFFUSION OF WATER THROUGH ISOLATED HUMAN EPIDERMIS

By F. RAY BETTLEY and ELAINE DONOGHUE
Institute of Clinical Research, Middlesex Hospital, London

THERE is reason for supposing that soap and other detergents increase the permeability of the skin. Thus, Kvorning and Svendsen[1] found that if 1 per cent 'Teepol' is incorporated with the test solution, patients sensitive to chromium and nickel react to a lower concentration than if chromium or nickel salts are used alone. Skog[2] found that pretreatment of the skin with soap or non-soap detergent led to a higher proportion of irritant reactions from dinitrochlorbenzol. Burckhardt[3] pretreated the skin with soap and found that the damage so inflicted predisposed to the experimental development of nickel sensitivity; but in his experiments gross damage was inflicted on the skin by the alkaline soap. This was perhaps more comparable with the increased sensitization to dinitrochlorbenzol obtained by Rockwell[4] in skin which was prepared by sand-papering.

The barrier which prevents passage between the tissues and the outside has been much investigated and is reviewed by Rothman[5]. It lies between the corneous layer and the Malpighian layer and is not identifiable with histological precision. According to Rothman, it is an electric double layer with hydrogen

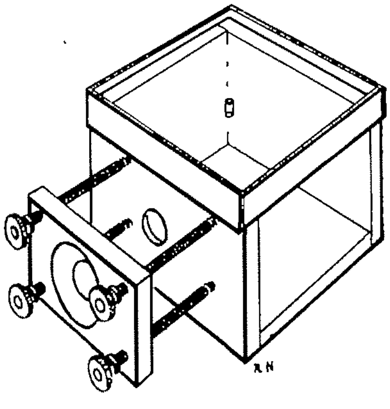


Fig. 1

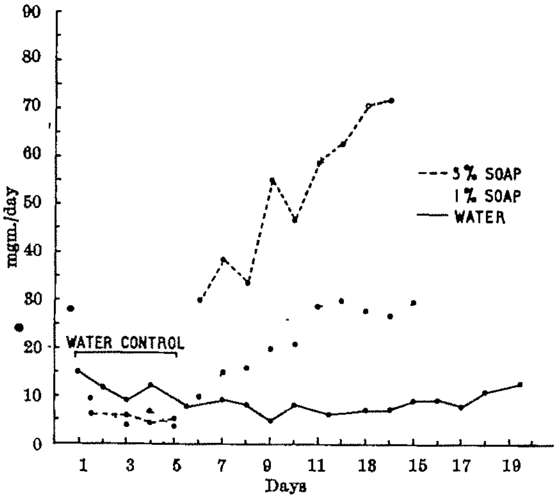


Fig. 2

ions on one side and hydroxyl on the other. It is impermeable or nearly so to water and to electrolytes; but allows the passage of lipoid-soluble substances. In the present studies isolated epidermis has been used for measuring the diffusion of water across this barrier layer.

The diffusion of water through isolated skin has been studied by various techniques. Burch and Winsor[6] and Berenson and Burch[7] used full-thickness skin which they placed over the end of a brass cylinder containing wet cotton wool. They found that the rate of water diffusion depended upon temperature and humidity, but was fairly constant over a temperature-range of 15–25° C. and of humidity between 20 and 60 per cent. Winsor and Burch[8] also measured water diffusion through the skin *in vivo*; they sealed a brass chamber to the skin of the abdomen and measured the amount of water vapour given off to dry nitrogen which was circulated through the chamber. It is important to note that they found the loss of water through intact living skin to be the same as that through dead skin measured by their brass cylinder method. This presumably indicates that there is no vital process involved. Mali[9] reached a similar conclusion; he used epidermis which had been separated by the heat method of Baumberger *et al.*[10]. Sheets of epidermis were sealed over cups of water and the loss of weight estimated at varying temperatures and humidities. Mali found that fresh living skin removed at operation gave the same results as post-mortem skin which had been dead for as long as two or three months.

In all these experiments it has proved possible to obtain constant measurements of loss of weight which presumably represent a steady diffusion of water vapour across the epidermal barrier which is independent of sweating or any other vital process. The present experiments were designed to study the effect of soap and other cleansers on this barrier layer.

The skin used was taken from the anterior abdominal wall one or two days after death. Subcutaneous fat was scraped off and the skin placed on the bench with the epidermis downwards. A beaker containing water at 80° C. was placed on the skin for 60 sec. The water was then emptied and the beaker inverted with the skin still adhering to it. The epidermis could then be stripped easily.

Cells were made of 'Perspex' about 4 cm. cube (Fig. 1). In one side of this cell was a circular window 7 mm. in diameter with bevelled edges over which the epidermis was placed. This was secured by a 'Perspex' plate clamped into position and lightly smeared with 'Vaseline' so as to make a water-tight joint. The cell was then filled with water to a level that just covered the epidermal window. In order to prevent evaporation from the open surface of the water, a layer of liquid paraffin was floated on (liquid paraffin shows no appreciable loss from evaporation in the conditions of the experiment). The cell was covered with a lid with a small perforation so that the pressure inside the cell was the same as that outside the window.

The room temperature was constantly between 24° and 25° C.; humidity varied between 50 and 65 per cent.

The cell was weighed as nearly as possible every day. When a piece of sheet rubber was clamped over the window the weight remained constant. Loss of weight was assumed to be due to the passage of water or water vapour across the epidermal window.

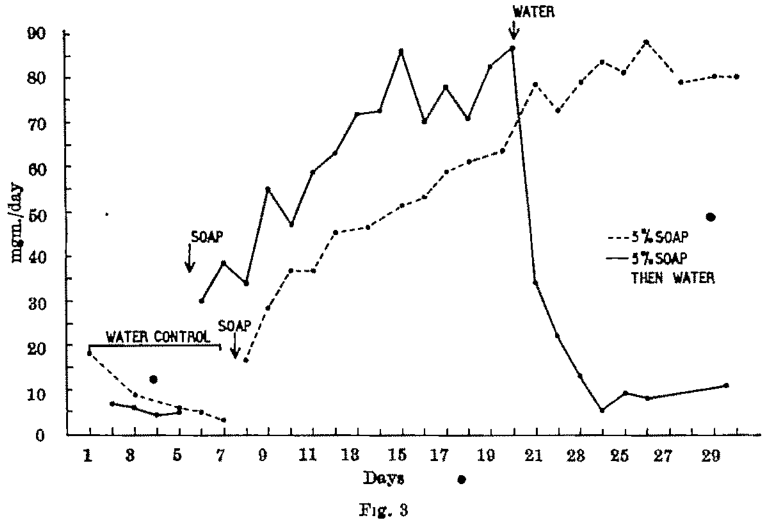


Fig. 3

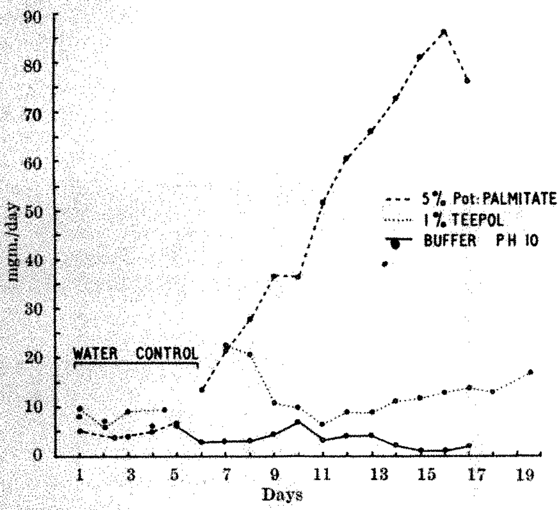


Fig. 4

In various experiments the water was replaced by aqueous solutions of soap, alkaline buffer and a non-soap detergent. Soap solution was made from the ordinary toilet soap provided for use in National Health Service hospitals in 1958. It is a toilet soap of good quality without free alkali or builders. 1 per cent and 5 per cent solutions were made with distilled water. Potassium palmitate was prepared from pure potassium hydroxide and palmitic acid in equimolecular proportions. 5 per cent solution was made with distilled water. 'Teepol' (Shell Chemical Co., Ltd.) is a soapless detergent consisting of secondary alkyl sulphate. 1 per cent and 5 per cent solutions were made with distilled water. Bile salt solution was made from purified ox bile salt, 1 per cent in distilled water. The buffer was the standard carbonate–bicarbonate buffer at *p*H 10.

In Figs. 2–4 are shown curves of individual experiments which are typical of the results in various groups.

When the cell contained tap water there was a steady loss in weight of about 5–10 mgm. daily. In prolonged experiments regular loss was continued over eighteen days during which the permeability of the epidermal membrane remained fairly constant (Fig. 2). After this, some preparations appeared to deteriorate in that loss of weight started to rise. It was thought that this might indicate disintegration of the epidermal barrier layer, and subsequent experiments were therefore usually limited to about that period. In the first day or two after the start of each experiment the initial loss was somewhat greater, perhaps because of a small amount of water remaining on the outside of the cell after the preparation had been set up. Small daily variations occurred and we cannot account for these. They were probably not attributable to changes in ambient temperature and humidity since when two cells were placed side by side in identical conditions they might show different variations, one losing more, one less than the daily average. Occasionally, also, an experiment gave unexpected results which could not be repeated and which were quite unlike other similar experiments. Some at least of these were probably due to errors which are not easy to recognize in any individual preparation. Such sources of error are minute damage to the epithelium which may occur particularly in relation to hair follicles during separation; imperfect sealing of the edges of the epithelial window; damage to the epithelium by the clamp which holds it in place; and contamination of the epithelium with paraffin. In all the subsequent experiments the behaviour of the epidermis with plain water was first observed for at least five days in order to make sure that it was normal.

The effect of putting soap solution into the cell was striking (Fig. 2). There was a rapid and progressive increase in loss of weight, which became more and more each day. With 5 per cent soap the daily loss of weight became constant after 8–10 days at a level of 70 mgm. or more, that is, ten times the rate of water; 1 per cent soap gave similar but less-marked effect, a maximum of 25–30 mgm. being reached in 5–7 days. The loss of weight remained at this high figure for at least several days, but when the soap solution was removed and replaced by water the rate of loss of weight returned rapidly to normal (Fig. 3). This presumably indicates that even after several days exposure to 5 per cent soap the epidermal barrier, though temporarily more permeable, is not essentially or permanently damaged. Similar results were obtained with 5 per cent potassium palmitate solution (Fig. 4).

It might be thought that the penetration of soap solution was due to its low surface tension allowing it to flow more readily through minute channels in the epidermis. We found, however, that if unglazed porcelain were placed over the window instead of epidermis the passage of soap solution was less rapid than that of water. Furthermore, when 1 per cent bile salt, which has a low surface tension, was put in the chamber the rate of diffusion through the epidermis remained about normal.

The presence of alkaline buffer at *p*H 10 in the chamber did not affect the rate of loss of weight (Fig. 4). 'Teepol' 1 and 5 per cent gave a loss of weight somewhat above that of water, up to about double, but not comparable in degree with the effect of soap (Fig. 3). These 'Teepol' solutions are slightly acid, but when buffered to about the same alkalinity as soap (*p*H 10) made no difference.

All these results were consistently repeatable in numerous experiments.

These experiments were carried out because we thought they might throw some light on the irritancy of soap solutions for the normal skin. Patch tests with soap and with a variety of other substances suggested that irritancy might be related in part to the power of soap to penetrate the epidermal barrier more efficiently than alkalis and other cleansers. The experiments recorded indicate that soap solutions have indeed the property of making the epidermis very much more permeable to water than is normal.

Winsor and Burch[8] found that the permeability of whole skin was so increased by scraping off the epidermis that the rate of loss of water was ten times that of intact skin, and was then the same as the rate of loss through pleura, heart muscle and other tissues. Comparing our results with these, it seems that the greatly increased permeability in contact with soap solutions represents virtually the complete loss of the epidermal barrier. This is far greater than the effect of 'Teepol' in a comparable detergent strength, suggesting that the cleansing or detergent property of soap and 'Teepol' is not the same as its penetrative effect. This is of interest for two reasons: First, because it lends some support to the suggestion that penetration of the epidermis rather than detergency may be important in the production of skin irritation since patch tests with 'Teepol' show far less irritation than soap; secondly, because this may throw some light on the nature of the epidermal barrier. Berenson and Burch[7] showed that treatment with lipoid solvents increased the permeability of the skin, from which it seemed that fat-soluble substances formed the essential part of the barrier. The action of a detergent, dissolving or emulsifying these fatty substances, might be supposed to be of a similar nature. Mali[9], in reviewing his own work and that of previous investigators, concluded that the barrier zone is a network of keratin fibres holding fatty substances in its meshes. This would explain the action of fat solvents and of detergents in destroying the barrier. Our experiments, however, indicate that 'Teepol' possesses this property much less than soap although it is a good detergent. More important still, we have observed that the effect of soap is reversible, and that the greatly increased permeability of the skin in contact with soap solutions returns to normal in a few days when soap is removed. This presumably indicates that the action of soap in increasing permeability cannot be due to its removing fatty or other substances from the barrier zone. Although we can draw no positive conclusions from our results, they seem to be more in conformity with Rothman's conception of a physico-chemical barrier. The barrier is made more permeable by soap without being permanently destroyed as it would be if fat solvency or detergency were responsible.

We are indebted to the Clinical Research Committee of the Middlesex Hospital for laboratory facilities and for a personal grant to one of us.

[1] Kvorning, S. A., and Svendsen, I. B., *J. Invest. Derm.*, **26**, 421 (1956).

[2] Skog, *Acta Derm.-venereol., Stockholm*, **38**, 1 (1958).

[3] Burckhardt, W., *Arch. Derm. Syph., Berlin*, **173**, 262 (1935).

[4] Rockwell, E. M., *J. Invest. Derm.*, **24**, 35 (1955).

[5] Rothman, S., *J. Lab. Clin. Med.*, **28**, 1305 (1943); "Physiology and Biochemistry of the Skin" (Chicago, 1954).

[6] Burch, G. E., and Winsor, T., *Arch. Derm. Syph., Chicago*, **53**, 39 (1946).

[7] Berenson, G. S., and Burch, G. E., *Amer. J. Trop. Med.*, **31**, 842 (1951).

[8] Winsor, T., and Burch, G. E., *Arch. Int. Med.*, **74**, 428 (1944).

[9] Mali, J. W. H., *J. Invest. Derm.*, **27**, 451 (1956).

[10] Baumberger *et al.*, *J. Nat. Cancer Inst.*, **2**, 413 (1941–42).

DEMYELINATION IN LAMBS BORN OF EWES MAINTAINED ON HIGH INTAKES OF SULPHATE AND MOLYBDATE

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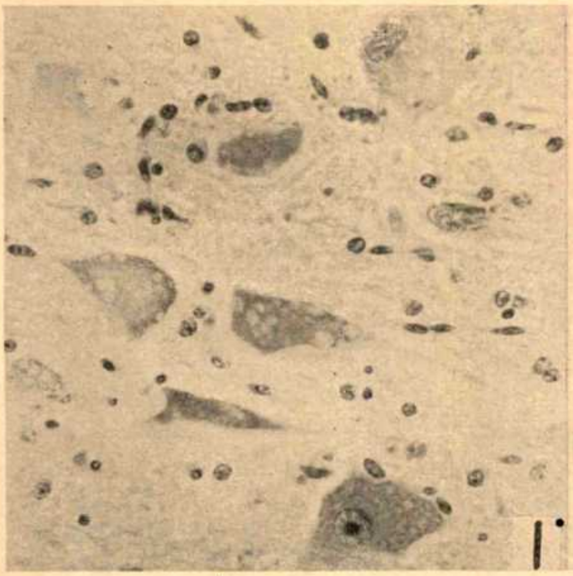
IN the course of studies to investigate the mechanisms underlying the interference with copper metabolism caused by high dietary intakes of molybdate and sulphate, the opportunity has been taken to examine the possible effects of such diets fed to pregnant ewes on the development of lambs after birth.

Eight two-year-old Cheviot ewes were divided into two groups one month after mating and fed the following diets daily: Group 1, 1 kgm. commercial grass cubes (copper content 5·0 μgm./gm.) supplemented with sodium sulphate to provide an intake of 10 gm. sulphate; group 2, 1 kgm. grass cubes supplemented as above and also with ammonium molybdate to give an intake of 50 mgm. molybdenum/day. This level of feeding was maintained to within one month of lambing, when the food offered was increased to 1·2 kgm./day. Tap water purified by



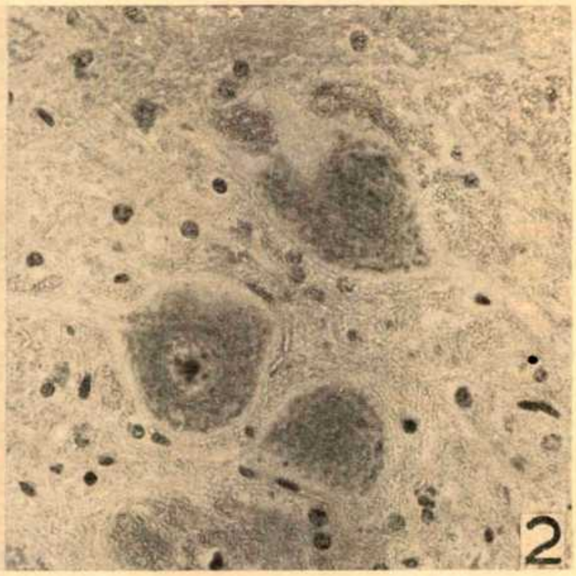
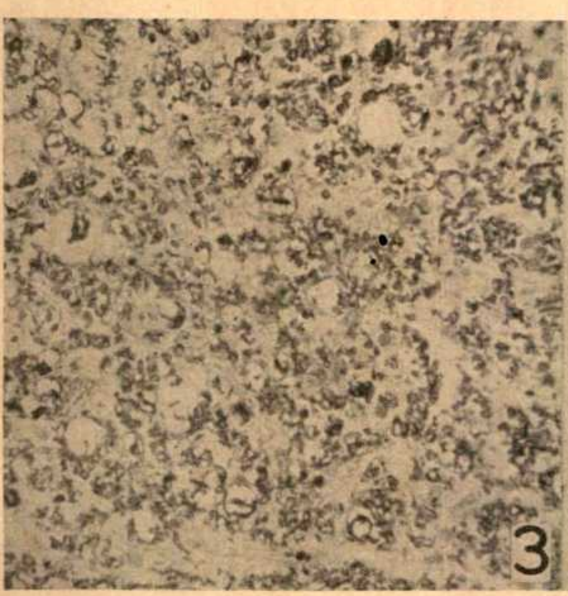


Fig. 1. Neurones in the red nucleus of a lamb in group 2 (high-sulphate and high-molybdate diet), showing chromatolysis and advanced cellular degeneration. Hæmatoxylin and eosin. (× 340)

Fig. 2. Neurones of normal appearance in the red nucleus of a lamb from group 1. (High-sulphate diet.) Hæmatoxylin and eosin. (× 340)



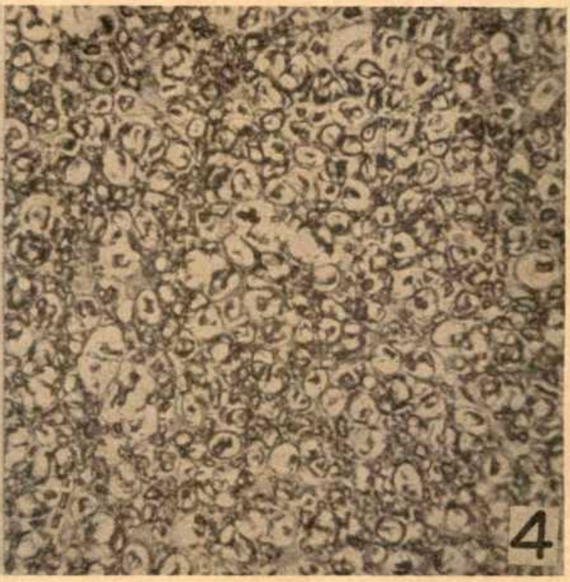


Fig. 3. Loss of myelin in a dorso-lateral column of white matter in the spinal cord of a lamb from group 2. (High-sulphate and high-molybdate diet.) Iron alum – hæmatoxylin. (× 280)

Fig. 4. Spinal cord of a lamb from group 1, showing absence of damage. This field corresponds with that shown in Fig. 3. Iron alum – hæmatoxylin. (× 280)

passage through an 'Elgastat Major Deioniser' (Elga Products, Ltd., London) was offered *ad lib.* in polythene buckets. One ewe of group 1 died after eighteen weeks on the experiment. All the remaining ewes lambed normally. Where twins were born, one animal was killed for liver-copper analysis immediately after birth. Inco-ordination of the hind limbs, progressing to affect both hind and fore limbs, appeared in one lamb of group 2 within three days of birth. Of the three remaining lambs in this group, a marked ataxia developed in one at eighteen days and another at sixty days of age.

Ataxic lambs were killed for autopsy and histological examination by being bled-out under pentobarbitone sodium anæsthesia. No macroscopic lesion was found during autopsy. Histological examination of the central nervous system was confined to the cerebral hemispheres posterior to the lateral cerebral fissure, the mesencephalon in the plane of the red nucleus and the cervical region of the spinal cord. Tissues were stained by Lillie's variant of the Weil–Weigert method for myelin, by the Marchi method for degenerating myelin and by hæmatoxylin and eosin. Lesions were recognized using all these preparations. There were degenerative changes consisting of chromatolysis progressing to loss of nuclei and all cytoplasmic detail in the large motor neurones of the red nucleus (Fig. 1). In one case such degeneration was present in the neurones of the ventral horns of grey matter in the spinal cord. Degenerative changes in the nuclei with shrinkage and loss of cytoplasm were present in the pyramidal cells of the cerebral cortex. The myelin of the cerebral sub-cortex was beaded or lost in diffuse, patchy areas, and in these areas the brain tissue was spongy with marked pericellular and perivascular spaces. The spinal cord in the neck showed a symmetrical diffuse demyelination in the dorso-lateral columns of white matter (Fig. 3) and in the ventro-lateral and ventral columns up to the median fissure. There was no 'cuffing' or cellular reaction suggestive of infection.

These lesions are closely similar to those described in field-cases of 'swayback' in lambs[1]. None of these degenerative changes was present in tissues prepared from lambs of group 1 (Figs. 2 and 4).

The results of copper determinations on livers from ewes and lambs are given in Table 1. The feeding of 10 gm. sulphate/day to ewes in group 1 resulted in the depletion of liver-copper stores to exceedingly low levels (mean : 8·8 μgm./gm. dry matter compared with a range of 186–1,374 μgm./gm. for 'normal' mature ewes and 7–106 μgm./gm. for copper-deficient ewes quoted by Cunningham[2]). Liver-copper levels of lambs in this group were low at birth and remained

Table 1. LIVER-COPPER OF EWES AND LAMBS ON DIETS CONTAINING SUPPLEMENTARY SULPHATE WITH AND WITHOUT ADDED MOLYBDENUM

Ewe No.	Treatment supplement	Liver copper (μgm./gm. dry matter)			Age at first signs of ataxia	Age at slaughter
		Ewe	Lamb killed at 1 day of age	Lamb killed when severely ataxic or at termination of exp.		
Group 1						
329	Sulphate (Group 1)	8·0	—	9·8	N*	100 days
56		11·6	10·8	8·1	N	103 days
38		8·3	8·9	9·5	N	87 days
28		7·3	(died before lambing, pregnancy toxæmia, twin lambs in uterus)			
Group 2						
181	Molybdenum sulphate (Group 2)	23·4	—	2·8	3 days	11 days
332		24·5	3·4	2·9	18 days	23 days
46		32·1	8·6	4·3	60 days	77 days
25		28·8	—	13·5	N	108 days

* N indicates no signs of ataxia up to termination of experiment.

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low afterwards (overall mean : 9·1 μgm./gm. compared with the 'normal' range of 74–430 μgm./gm., ref. 2), falling within the range reported for field-cases of copper-deficiency (4–34 μgm./gm., ref. 2). No case of ataxia was noted in lambs of this group, and growth and general condition in the period before slaughter remained excellent. This finding is not in line with the experiences of Marston[3], who concluded from field-experiments that ataxia is likely to develop in the lamb if the copper in the liver of the ewe falls to less than 20 μgm./gm. and becomes an almost invariable consequence if it is less than 10 μgm./gm.

Ewes receiving high levels of both sulphate and molybdenum in the diet (group 2) retained rather more copper in their livers (mean : 27·2 μgm./gm.) than the ewes of group 1, but were markedly inferior in physical condition and showed the characteristic loss of 'crimp' in wool associated with physiological copper-deficiency[4]. Allcroft and Lewis[5] noted a similar effect on liver-copper storage in the pregnant ewe from feeding molybdenum at 50 mgm./day. Analyses of lamb livers at the end of our experiment suggest that transfer of copper to the lamb was restricted by feeding molybdenum. Insufficient information is available to determine whether this is a result of diminished transfer to the fœtus or the result of decreased transfer during the suckling period ; but it is noteworthy that others[5] have failed to find any influence of feeding large quantities of molybdenum to the ewe on levels of copper in the liver of the new-born lamb.

The lambs in group 2 showing ataxia and degenerative changes in nervous tissue all had extremely low liver-copper values (mean : 3·3 μgm./gm.) and the only lamb in this group not so affected had an unaccountably high content of copper in its liver.

So far as we are aware, this experiment represents the first successful attempt to induce demyelination in the lamb by dietary treatment. Few details have as yet been published of previous attempts of other workers to induce this condition by feeding diets low in copper, but a survey of published results presenting liver-copper analyses from lambs suffering from 'swayback' or 'enzootic ataxia' under field-conditions[1,2,6–9] suggests that clinical symptoms and associated degenerative changes in the central nervous system may occur when the liver-copper of the lamb falls below about 20 μgm./gm. dry matter. It remains to be determined whether the absence of ataxia in the lambs of group 1, despite their very low levels of liver-copper, is due to the fact that no known metabolic antagonist of copper was present in the diet in quantity to impede the utilization of these small stores. Without postulating the existence of such a restrictive agent in some diets, it is difficult to understand why ataxia can occur under field-conditions at concentrations of liver-copper considerably higher than those found necessary in the present investigation.

[1] Innes, J. R. M., and Shearer, G. D., *J. Comp. Path.*, **53**, 1 (1940).
[2] Cunningham, I. J., *N.Z. J. Sci. Tech.*, **27** (A), 381 (1946).
[3] Marston, H. R., *Physiol. Rev.*, **32**, 66 (1952).
[4] Lee, H. J., *J. Agric. Sci.*, **47**, 281 (1956).
[5] Allcroft, R., and Lewis, G., *Landbouwk. Tijdschr.*, **68**, 711 (1956).
[6] Bennetts, H. W., and Beck, A. B., *Austral. C.S.I.R. Bull.*, No. 47, 30.
[7] Shearer, G. D., and McDougall, E. I., *J. Agric. Sci.*, **34**, 207 (1944).
[8] Spaïs, A., "A Contribution to the Study of Enzootic Ataxia in Sheep in Greece", thesis, Univ. of Thessaloniki (1956).
[9] Howell, J. McC., and Davison, A. N., *Biochem. J.*, **72**, 365 (1959).

EFFECT OF STREPTOMYCIN ON *ESCHERICHIA COLI*

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Damage by Streptomycin to the Cell Membrane of *Escherichia coli*

It has been shown in this laboratory that streptomycin causes a sensitive strain (but not a streptomycin-resistant strain) of *Escherichia coli* to excrete certain 5′-ribonucleotides at a relatively high rate[1]. It seemed possible that this effect might be due to interference with nucleic acid formation, resulting in an increase in the intracellular pool of free nucleotides. However, nucleotides (or their derivatives) exert a feedback effect on their own biosynthesis, as shown by the fact that an excess of the purine or the pyrimidine required by certain mutants inhibits accumulation of precursors[2]. If these feedback mechanisms effectively regulate nucleotide biosynthesis, a simple block in nucleic acid formation would not be expected to cause an increase in intracellular nucleotide levels. Alternatively, nucleotide excretion might be due to a breakdown of the permeability barrier that normally retains these compounds within the cell. This mechanism would not require elevation of the intracellular levels.

In order to distinguish between these two mechanisms, the levels of nucleotides have been compared in the intracellular pool and in the external medium during exposure to streptomycin. The comparison has also been extended to amino-acids.

For these studies streptomycin (60 μgm./ml.) was added to an aerobic culture of *E. coli* strain *W* growing exponentially in mineral-glucose medium[3] at about 4 × 10[8] cells/ml. Under these conditions the lethal action of streptomycin becomes rapid after a delay of 5–10 min. Cells were harvested after 20 min., when nucleotide excretion was just beginning, and after 45 min., which corresponds to roughly half-maximal excretion. The culture filtrate was harvested after 60 min., when excretion was approaching completion.

Table 1 presents the values, averaged from 4 experiments, for those nucleotides that are excreted heavily. It is seen that after 20 min. of streptomycin treatment the intracellular levels were essentially unchanged, except for an unexplained rise in uridine monophosphate. (There was also a rise in a minor unidentified diphosphate.) At 45 min., when excretion was well under way, all the nucleotides showed a decrease, rather than a rise, in intracellular level, thus pointing to loss of integrity of a permeability barrier. The effect on nucleotide excretion was irreversible, persisting in cells transferred to streptomycin-free medium.

The various mononucleotides showed rather similar extracellular concentrations after streptomycin treatment, despite appreciable differences in their intracel-

Table 1. EFFECT OF STREPTOMYCIN ON NUCLEOTIDE EXCRETION AND ON INTRACELLULAR LEVELS

Compound	Nucleotide concentration (moles × 10^{-16}/ml. of culture)			
	Intracellular*			Extracellular†
	Untreated	SM 20 min.	SM 45 min.	SM 60 min.
AMP	3·1	3·1	2·1	39·2
GMP	1·4	1·8	1·0	39·1
UMP	2·0	4·4	4·2	26·8
CMP	< 0·4	< 0·4	< 0·4	27·6
ADP	2·1	2·3	2·3	14·2
ATP	2·6	2·4	2·0	8·6
DPN	4·1	3·9	2·4	5·0

Nucleotides were isolated from the culture filtrate by adsorption on charcoal and elution by alcoholic ammonia, and from the cells by treatment with 5 per cent trichloracetic acid, which was then removed by extraction with ether. The individual nucleotides were separated on a column of 'Dowex 1-x8' formate by elution with a linear gradient of 5 *N* formic acid, followed by a gradient of 0·8 *M* ammonium formate in 5 *N* formic acid. Concentrations were estimated spectrophotometrically.

* The concentrations per ml. of intracellular water are about 1,500 times the intracellular values reported here per ml. of culture. The excretion thus represents leakage down a steep concentration gradient.

† The extracellular nucleotide concentrations in untreated cultures were too low to be measured.

SM, streptomycin.

lular concentrations. Larger compounds (diphosphopyridine nucleotide, adenosine diphosphate and adenosine triphosphate) were also excreted, but in smaller proportion to the intracellular concentration. It is seen that the adenosine monophosphate excreted in 60 min. amounted to about 12 times the initial intracellular content. This finding fits the isotopic evidence[1] that the bulk of the excreted nucleotide is synthesized *de novo* during streptomycin treatment.

Since amino-acids also form a substantial intracellular pool, any general loss of integrity of the cell membrane should increase their excretion as well. Indeed, though amino-acids, in contrast to nucleotides, were found to be excreted appreciably even by normal cells (cf. ref. 4), streptomycin treatment for 50 min. caused a four-fold increase in their excretion. This treatment also caused an even greater drop in intracellular levels of amino-acids than had been observed for nucleotides.

Further evidence on the nature of streptomycin action was provided by the observation that its addition to a growing culture progressively destroyed the capacity of the cells to concentrate valine labelled with carbon-14. This treatment also increased the metabolism of added labelled citrate by the cells, which were initially relatively impermeable to this compound. On the basis of all the above findings, it is concluded that streptomycin damages the permeability barrier to small molecules.

Attention has also been given to the conditions required for streptomycin action. Chloramphenicol, which blocks protein synthesis[5], has been found to block the action of streptomycin. With streptomycin alone, under the conditions described above, net formation of protein halted after about 15 min., with an increment of 10–15 per cent; formation of ribo- and deoxyribo-nucleic acid persisted longer and reached increments of 40 and 55 per cent, respectively. With chloramphenicol (20 μgm./ml.) plus streptomycin, as with chloramphenicol alone[5], no protein synthesis was detectable, while ribonucleic acid increased by 70 per cent. Furthermore, chloramphenicol completely blocked both the bactericidal action of streptomycin and its effect on nucleotide excretion. Similar protection against these last two actions had also been obtained by depriving certain auxotrophic mutants of the required amino-acid or pyrimidine (Davis, B. D., and Roth, H., unpublished observations). These results indicate that a certain amount of protein synthesis is required for streptomycin action. They also show that the effects of streptomycin on metabolite excretion and on viability are not due simply to cessation of growth, or to the unbalanced formation of nucleic acid without protein.

It is clear that streptomycin, under appropriate conditions, alters a permeability barrier. Since this effect increases the outward permeability to nucleotides and to amino-acids, and the inward permeability to citrate and (following communication) to streptomycin itself, it seems reasonable to infer that the permeability barrier involved is the cell membrane. This action of streptomycin is reminiscent of that of detergents[6] and polymyxin[7], the attack of which on the membrane also results in metabolite excretion and cell death. However, the facts reported above imply that streptomycin does not directly damage the existing membrane, but rather causes defects in membrane formed in its presence by streptomycin-sensitive cells. The action of streptomycin on membrane formation is thus analogous to that of penicillin on bacterial wall formation[8]. The following communication discusses the significance of this action for the bactericidal effect of streptomycin.

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[1] Roth, H., Amos, H., and Davis, B. D., *Bact. Proc.*, 131 (1959); *Biochim. Biophys. Acta* (in the press).

[2] Brooke, M. S., Ushiba, D., and Magasanik, B., *J. Bact.*, 68, 534 (1954). Yates, R. A., and Pardee, A. B., *J. Biol. Chem.*, 221, 757 ~~193~~ (1956). Gots, J. S., *Arch. Biochem.*, 29, 222 (1950). Gots, J. S., and Goldstein, J., *Science*, 130, 623 (1959).

[3] Davis, B. D., and Mingioli, E. S., *J. Bact.*, 60, 17 (1950).

[4] Dagley, S., and Johnson, A. R., *Biochim. Biophys. Acta*, 21, 270 (1956).

[5] Gale, E. F., and Folkes, J. P., *Biochem. J.*, 53, 493 (1953). Wisseman, C. L., Smadel, J. E., Hahn, F. E., and Hopps, H. E., *J. Bact.*, 67, 662 (1954).

[6] Hotchkiss, R. D., *Ann. N.Y. Acad. Sci.*, 46, 479 (1946). Salton, M. R. J., *J. Gen. Microbiol.*, 5, 391 (1951).

[7] Newton, B. A., *J. Gen. Microbiol.*, 9, 54 (1953); *Bact. Rev.*, 20, 14 (1956).

[8] Lederberg, J., *J. Bact.*, 73, 144 (1957). Park, J. T., and Strominger, J. L., *Science*, 125, 99 (1957).

Uptake of Streptomycin by *Escherichia coli*

THE present communication will attempt to relate the findings of the previous communication to the kinetics and the sites of streptomycin uptake by the cells.

For this purpose streptomycin labelled with carbon-14 was added to a growing culture. 1-ml. samples were taken at intervals and the cells were recovered on a 'Millipore' filter and washed with 3 × 5 ml. of water to remove the extracellular fluid. Further washing removed no radioactivity. In contrast, washing with buffer removed 99 per cent of the radioactivity from early samples and 94 per cent from late samples, indicating that most of the streptomycin, even after it had killed the cells, was held by ionic linkage.

Results of a typical experiment are presented in curve *SM* of Fig. 1. The initial uptake of streptomycin was extremely rapid, being essentially complete by the time the first sample could be taken and filtered (about 1 min.). During the next 15 min. the cell content remained constant. There was then a secondary rise, levelling off after an hour at a three- to four-fold increase. This rise began at about the time (previous communication) when excretion of

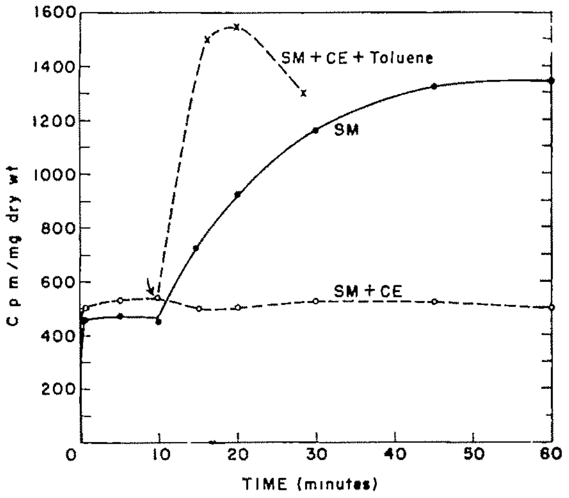

Fig. 1. Uptake of streptomycin labelled with carbon-14. 40 μgm./ml. of labelled streptomycin (formed from uniformly labelled glucose) was added to an exponentially growing shaking culture in minimal medium, and to two similar cultures (labelled *CE*) to which chloramphenicol (20 μgm./ml.) had been added 2 min. earlier. At the time indicated by the arrow, one *CE* culture received 0·02 ml. of toluene per ml. of culture

intracellular metabolites began and protein synthesis ceased.

We initially attempted to explain these observations on the basis of the assumption that the cell is freely permeable to streptomycin. Part of the initial uptake would then occur at intracellular sites where streptomycin would interfere with normal protein (cf. ref. 1) and nucleic acid synthesis. Resulting defective formation of protein and perhaps other membrane constituents could explain the observed damage to the permeability barrier of the cell.

The kinetics of uptake of streptomycin, however, is difficult to interpret on this basis. Not only is a plateau reached with streptomycin much more rapidly than has been observed with many added substances which are known to penetrate (for example, amino-acids), but at 0° the initial uptake was found to be as high as at 37°, and was still too rapid to follow. Furthermore, the observation of a secondary uptake would require the postulate, in a freely permeable cell, of the formation of added receptors for streptomycin. Since streptomycin complexes readily with free nucleic acids *in vitro*[2], it seemed possible that these receptors might be the excess ribo- and deoxyribo-nucleic acids which are formed in the presence of streptomycin after protein formation ceases (see previous communication). However, though the addition of chloramphenicol, in the presence of streptomycin, causes an even more pronounced synthesis of excess nucleic acids (previous communication), this addition completely prevented the secondary uptake of streptomycin (curve *SM* + *CE* of Fig. 1). It would therefore be necessary to postulate the formation of other, unknown receptors, in a quantity several times that initially available, and this explanation of the secondary uptake seems rather improbable.

Alternatively, it seemed possible that the cell might be impermeable to streptomycin, the rapid initial attachment of which would then be restricted to the cell wall and the external surface of the cell membrane. Some of its attachments on the latter locus would be responsible for the observed formation of defective membrane. This membrane damage would then permit entry of streptomycin into the cell, where it would be bound by nucleic acids[2] and possibly other anionic constituents. The block in protein synthesis, and later in nucleic acid synthesis, would follow the entry of streptomycin into the cell.

To distinguish between these two mechanisms, observations were made on the response of streptomycin uptake to known damage to the membrane. In an experiment with cells the uptake of which was being held at the initial level by the presence of chloramphenicol, the addition of toluene resulted in a large and rapid further uptake (Fig. 1). In other experiments cells were treated with toluene before exposure to streptomycin. This procedure resulted in a several-fold increase in the initial uptake; and similar treatment with polymyxin *B* (cf. ref. 3) had an even greater effect.

These observations indicate that during action of streptomycin its initial uptake, at least within measurable limits, occurs only outside the cell membrane, and the secondary uptake depends on subsequent damage to the membrane. It therefore appears likely that the growing membrane, known to be damaged by streptomycin, is the primary site of action of the drug. It has not yet been possible to determine whether the damage to the membrane is a sufficient as well as a necessary cause of the bactericidal effect of streptomycin, or whether this effect depends on penetration of the drug.

A resistant mutant exhibited the same initial uptake of streptomycin as its sensitive parent, but no secondary uptake. It therefore appears that in this case resistance depends on an alteration in the membrane-forming apparatus such as to prevent streptomycin from damaging the membrane. This mechanism of resistance would explain the observation that in a diploid strain of *E. coli* streptomycin sensitivity is dominant to resistance[4]. The membrane in such a cell would presumably contain both sensitive and resistant sites, and streptomycin action on the sensitive sites would have the same consequences as in a fully sensitive cell.

The mechanism of streptomycin action proposed here provides an explanation for the well-known and puzzling synergism observed between streptomycin and other bactericidal agents. A drug that damages the permeability barrier of a cell would promote the entry of other drugs.

This work will be published in detail. It has been supported by Grants *E*-2012 of the U.S. Public Health Service and *G*-9078 of the National Science Foundation. We are grateful to Merck Sharp and Dohme for a generous gift of radioactive streptomycin, and to Miss Elizabeth Vollmayer for excellent technical assistance.

Some early experiments were performed by two of us (A. K. A. and B. D. D.) at the Department of Pharmacology, New York University College of Medicine.

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[1] Erdos, T., and Ullmann, A., *Nature*, 183, 618 (1959).

[2] Cohen, S. S., *J. Biol. Chem.*, 166, 393 (1946).

[3] Newton, B. A., *J. Gen. Microbiol.*, 9, 54 (1953); *Bact. Rev.*, 20, 14 (1956).

[4] Lederberg, J., *J. Bact.*, 61, 549 (1951).

COMMERCIAL APPLICATION OF GIBBERELLIC ACID TO HOPS

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THE effect of gibberellic acid as a growth-promoting substance caused attention to be focused upon it, when a great disadvantage of newly bred triploid hop plants became apparent. The triploid material, being partly sterile, tended to hang in the burr stage for a long period, and it was considered that gibberellic acid could perhaps force the triploids from the burr stage, besides giving an economic increase in yield.

To implement such a hypothesis, preliminary applications of gibberellic acid were carried out to determine the optimal time of application. For these determinations, the potassium salt of gibberellic acid under the trade name 'Gibrel' was used. Laterals at different stages of burr development on different plants of the triploid material, known as 12/5/28, were completely covered in a plastic bag and sprayed by a Rega continuous-spray unit with the 'Gibrel' solution of concentration 50 parts per million. These preliminary sprays showed that the time of application could reasonably be narrowed down to when the stigmas of the female flowers were ⅛ in. or greater in length. Earlier application of the spray resulted in excessive elongation of the sub-laterals (this was of the order of 100 per cent in comparison with the control laterals) and later application showed a decrease in overall effect.

Once the time of application had been suitably determined, a further trial with different strengths of application was attempted. Concentrations of 50, 25, 12·5, 6·25 and 0·625 parts per million (p.p.m.) respectively were used, with applications carried out exactly as above on laterals of different plants with individual cone weights for each treatment being recorded. These were compared to the cone weights from similar laterals from unsprayed vines used as controls. The gibberellic acid-sprayed material showed typical reactions[1]. The leaves became paler within 4 days; this, however, was only temporary and after a further 6 days the leaves had regained their normal colour. The sprayed material showed normal development of the cones, with no abnormal growth of the sub-laterals, and was approximately 10 days in advance in maturity on the unsprayed control material. Not only was the maturity of the cones pushed forward, but also the cones along the sprayed laterals were more even in development, so that the harvest time became more critical, with practically all cones at the same stage of maturity at the same time. However, those cones developed from gibberellic acid-sprayed burr were more subject to wind burn than the controls and the degree of burn was approximately the same for all concentrations. The most significant part of the growth of the gibberellic acid material was the great initial surge of development, which although slowing down with time, was still sufficient to ripen the cones 10 days before the control cones. The actual cone weights are set out in Table 1.

Table 1

Concentration of spray (p.p.m.)	No. of cones in treatment	Weight/cone (gm.)
0·625	164	0·82
6·25	212	0·85
12·5	225	1·10
25·0	148	1·40
50·0	151	1·44
Control	594	0·77

It was decided that the concentration of gibberellic acid at 12·5 p.p.m. would give the desired effect of increase in yield and decrease in maturity time, without being too expensive for an equitable return.

Since most commercial growers of hops are equipped with overhead sprays, a full plant trial was adopted whereby the gibberellic acid applied at 12·5 p.p.m. would be sprayed from above the plant by means of a fine nozzle shower-head. The variety used was a locally bred, high analysis hop called Pride of Ringwood. The shower-head was connected to a carbon dioxide cylinder with an apparent head pressure of 30 lb./sq in., but due to the friction loss along the long, small diameter hose necessary to reach the tall crop, the actual spray was delivered at somewhat less than 30 lb./sq. in.; however, excellent coverage of the plants was effected due to the fine spray achieved. The time of application was allowed to progress a little past the optimal time to make sure that most of the burr was at a receptive stage. The average yield per sprayed hill was 7·25 lb. while the average yield for the control hills was 5·20 lb. This gave an average increase of 2·05 lb./hill—a large increase when considered in terms of 850 hills/acre. On yield results alone, the application of gibberellic acid seemed justifiable, since the yields were increased and the decreased time of maturity would allow for a less restrictive harvest period. The main interest in bought hops is the percentage of α resin in the purchased product. The two lots of material, treated with gibberellic acid and controls, were analysed for α-resin content by the Verzele rapid polarimetric method[2], and further tests for total soft resin content were carried out by the Walker–Hastings method[3].

Table 2

	α-Resin (per cent)	Total soft resin (per cent)
Treated hops	1·8	9·7
Untreated hops	10·16	21·8

Table 2 shows that the most important substance in the hops from the brewer's point of view has been greatly decreased by application of gibberellic acid. Further work on estimation of oils by gas chromatography was carried out and appears to indicate that here again there is a difference in percentages of the various oil components between gibberellic acid-treated and normal hops; however, this work is inconclusive and requires verification.

[1] Brian, P. W., and Grove, J. F., *Endeavour* (July 1957).

[2] Wallenstein Lab. Comm. No. 68, **20**, 7 (1957).

[3] *J. Inst. Brew.*, **39**, 509 (1933).

LETTERS TO THE EDITORS

PHYSICS

Growth of the Tritium Content of Atmospheric Molecular Hydrogen

A SERIES of measurements of the abundance of tritium in atmospheric molecular hydrogen, covering the period 1956–58, were carried out by Gonsior in Germany[1]. These measurements were interpreted by him as showing that over that period the abundance was increasing linearly by 5 × 10^4 T.U. a year commencing with the 'Castle' series of thermonuclear weapon tests in the spring of 1954. (1 T.U. is equivalent to 1 tritium atom in 10^{18} hydrogen atoms.)

We, too, believe that the growth must be due to atomic energy operations; but in our view Gonsior has taken insufficient account of abundances reported directly or otherwise at least four times prior to the end of 1958:

(*a*) Faltings and Harteck[2-5] measured the tritium abundance in hydrogen from Hamburg air collected in 1948–49. They quoted the abundance as being $0{\cdot}38 \pm 0{\cdot}125 \times 10^4$ T.U.

(*b*) Grosse, Kirshenbaum, Kulp and Broecker[6] reported $1{\cdot}66 \pm 0{\cdot}097 \times 10^4$ T.U., having examined air collected at Buffalo, N.Y., in the spring of 1952.

(*c*) Begemann[7] stated that about 1 per cent of the world inventory of natural tritium was present in gaseous hydrogen during the spring of 1954. As he believed at the time that natural tritium totalled 1·8 kgm., we may suppose that the abundance in hydrogen was then about $3{\cdot}4 \times 10^4$ T.U. in the United States.

(*d*) Craig[8] reported that a dozen samples of air examined by Begemann had abundances ranging from 5 to 10 × 10^4 T.U., averaging about 8 × 10^4 T.U. These determinations were described as 'recent' in November 1956[9]. The origin of the samples was not stated.

These results, together with those of Gonsior, are presented in Fig. 1, the ordinate scale of which is logarithmic. As can be seen, the data, with the possible exception of Gonsior's first Nuremberg result, are consistent with an abundance of tritium in northern hemisphere tropospheric hydrogen which was increasing not only before 1954 but also before the first reported detonation of any 'hydrogen device', in October–November 1952. It furthermore appeared to us that the abundance could be empirically represented as increasing exponentially with a doubling time of 18 months, as is shown by the line drawn through the experimental points in Fig. 1.

In order to establish whether the tritium abundance was continuing to increase in the manner suggested, we measured the tritium activity in mixtures of atmospheric hydrogen and neon collected at the end of 1958 and in the summer of 1959, the gas mixtures being extracted from the helium–hydrogen–neon fraction that arises in commercial liquid air distillation by fractional desorption from charcoal. We have no reason to suppose that isotopic fractionation of tritium from hydrogen occurs either in the distillation or in the desorption process. The hydrogen–neon mixtures each came from several distillation plants situated in the United Kingdom, and each sample represented an averaging period of two or three weeks. The mixtures were counted in a Geiger–Muller counter using di-ethyl ether as a quenching agent, the percentage of hydrogen in the mixture being first determined by mass-spectrometric means. For air over the United Kingdom we deduced tritium/hydrogen = $38{\cdot}1 \pm 1 \times 10^4$ T.U. (November–December 1958) and $36{\cdot}5 \pm 1 \times 10^4$ T.U. (December 1958). The following summer, measurement showed tritium/hydrogen = $48{\cdot}6 \pm 1{\cdot}5 \times 10^4$ T.U. (July–August 1959). The errors quoted are in each case those arising from uncertainty in the volume of gas counted together with the statistical counting error. These results are shown in Fig. 1, the growth-rate for the first half of 1959 being 18 × 10^4 T.U. per year (mean), three and a half times the mean rate measured earlier by Gonsior.

We have shown, therefore, that the abundance of tritium in atmospheric hydrogen is still increasing in a manner not inconsistent with an 18-month doubling time, but are unable to advance any satis-

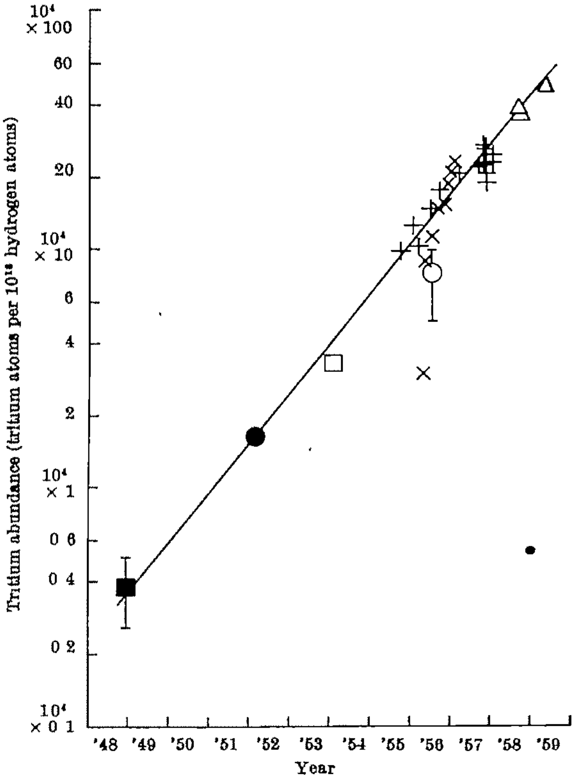

Fig. 1 Increase of tritium in the northern hemisphere atmospheric hydrogen during the last decade. ■, Faltings and Harteck; ●, Grosse *et al.*; □, Begemann; ○, Begemann (Craig); +, Gonsior–Hamburg; ×, Gonsior–Nuremberg; △, own measurements

factory explanation of this phenomenon, which could well be fortuitous, depending on the extent to which weapons trials held prior to October 1952 released tritium to the atmosphere, and on the extent to which artificial tritium escapes to the atmosphere in the course of its production. Were this tritium to be distributed uniformly throughout the hydrogen of the atmosphere, a matter which we doubt, then the world integral of tritium in this form would be about 2 megacuries, that is, about 200 gm., at the present time. Whatever factors one allows for non-uniform distribution in the atmosphere the integral is still probably much less than the hundreds of megacuries of artificial tritium that we know to be present in the water bodies of the Earth[10,11].

We are grateful to Mr. B. Bann of British Oxygen Gases, Ltd., Wembley, Middlesex, for supplying us with the gas samples and to Mr. J. Pennington of the same Company for having separated them from their associated helium.

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[1] Gonsior, B., *Naturwiss.*, 46, 201 (1959).
[2] Faltings, V., and Harteck, P., *Z. Naturforsch.*, 5, a, 438 (1950).
[3] Harteck, P., and Faltings, V., *Nature*, 166, 1109 (1950).
[4] Faltings, V., *Naturwiss.*, 40, 409 (1953).
[5] Harteck, P., *J. Chem. Phys.*, 22, 1746 (1954).
[6] Grosse, A. V., Kirshenbaum, A. D., Kulp, J. L., and Broecker, W. S., *Phys. Rev.*, 93, 250 (1954).
[7] Begemann, F., "Nuclear Processes in Geologic Settings" N.A.S.–N.R.C. Publication 400, 166 (1956).
[8] Craig, H., "The Effects of Atomic Radiation on Oceanography and Fisheries", N.A.S.–N.R.C. Publication 551, 103 (1957).
[9] Craig, H., *Phys. Rev.*, 105, 1125 (1957).
[10] Begemann, F., and Libby, W. F., *Geochim. and Cosmochim. Acta*, 12, 277 (1957).
[11] Bolin, B., Second U.N. International Conference on the Peaceful Uses of Atomic Energy, *A/CONF* 15/*P*/176 (1958).

Horizontal Drifts in the *E*-Region at Waltair

A CONSIDERABLE amount of work has been done on the study of horizontal drifts in the *E*-layer at high-latitude stations, the results of which were summarized by Briggs and Spencer[1]. As there is practically no work done on this aspect at low-latitude stations, systematic measurements of *E*-region drifts were undertaken at Waltair (geogr. lat. 17° 43′ N., geo.-mag. lat. 7·4°) from the beginning of the International Geophysical Year.

The results presented here are based on the analysis of data obtained at Waltair for two years, June 1957–May 1959. The similar fades method of Mitra was used with a slightly modified technique of recording[2]. Measurements were made on a frequency of 2·5 Mc./sec. for all the 24 hr., drift records using the reflexions from E_s only being considered during night-time. Observations were taken at hourly intervals on all the world days and, in addition, measurements were made on at least three more days in each month so that the data were available for at least six days in each month. The drift speed and direction are calculated by the similar fades method. Following the usual practice, the north–south and east–west components of the observations taken during each month and season are plotted separately and mean curves drawn through the points as judged by the eye. These curves were subjected to harmonic analysis to obtain steady 24-hr. and 12-hr. components. In this communication, only the results of harmonic analysis of the data for the four seasons taken during the two years are presented Table 1 gives the results of the harmonic analyses thus obtained.

Table 1

Summer	1957	N.–S.	$-37\cdot7 + 9\cdot0 \sin(t+291^\circ) + 30\cdot0 \sin(2t+110^\circ)$
		E.–W.	$+1\cdot5 + 79\cdot0 \sin(t+46^\circ) + 19\cdot0 \sin(2t+342^\circ)$
Summer	1958	N.–S.	$-46\cdot0 + 26\cdot0 \sin(t+314^\circ) + 32\cdot3 \sin(2t+99^\circ)$
		E.–W.	$+12\cdot0 + 74\cdot0 \sin(t+34^\circ) + 2\cdot7 \sin(2t+81^\circ)$
Autumn	1957	N.–S.	$-33\cdot3 + 8\cdot0 \sin(t+199^\circ) + 31\cdot0 \sin(2t+130^\circ)$
		E.–W.	$-2\cdot2 + 67\cdot0 \sin(t+37^\circ) + 29\cdot4 \sin(2t+329^\circ)$
Autumn	1958	N.–S.	$-19\cdot0 + 20\cdot0 \sin(t+140^\circ) + 47\cdot5 \sin(2t+101^\circ)$
		E.–W.	$+5\cdot4 + 99\cdot0 \sin(t+39^\circ) + 2\cdot4 \sin(2t+352^\circ)$
Winter	1957–1958	N.–S.	$-12\cdot9 + 45\cdot0 \sin(t+85^\circ) + 32\cdot0 \sin(2t+40^\circ)$
		E.–W.	$+4\cdot0 + 76\cdot0 \sin(t+41^\circ) + 15\cdot5 \sin(2t+346^\circ)$
Winter	1958–1959	N.–S.	$-7\cdot0 + 42\cdot0 \sin(t+92^\circ) + 30\cdot0 \sin(2t+41^\circ)$
		E.–W.	$+3\cdot2 + 78\cdot0 \sin(t+42^\circ) + 21\cdot3 \sin(2t+346^\circ)$
Spring	1958	N.–S.	$-20\cdot1 + 30\cdot5 \sin(t+136^\circ) + 41\cdot0 \sin(2t+60^\circ)$
		E.–W.	$+1\cdot1 + 71\cdot0 \sin(t+57^\circ) + 22\cdot0 \sin(2t+344^\circ)$
Spring	1959	N.–S.	$-24\cdot0 + 42\cdot0 \sin(t+99^\circ) + 18\cdot8 \sin(2t+84^\circ)$
		E.–W.	$+8\cdot2 + 78\cdot5 \sin(t+42^\circ) + 11\cdot6 \sin(2t+1^\circ)$

Typical curves showing the diurnal variation of the north–south and east–west components for the summer and winter seasons of 1958–59 are shown in Fig. 1.

The main features of the *E*-region drifts at Waltair may be summarized as follows. The 24-hourly component is the predominant periodic component of the drift vector, while the steady and 12-hourly components are nearly of the same order of magnitude. The direction of the steady component, which is generally towards south or south–south-east, does not show any systematic change from season to season, while the magnitude shows a seasonal variation with a minimum in winter and a maximum in summer. The amplitude of the 24-hr. periodic north–south component is always less than the east–west component, while the 12-hourly periodic north–south component is always higher than the east–west component, the difference being particularly marked in summer and autumn seasons. The phase difference between north–south and east–west components is such that the drift vector rotates clockwise in all seasons for the 12-hourly periodic component and also for the 24-hourly periodic component except the summer season for which an anti-clockwise sense of rotation is definitely observed. The phase difference between north–south and east–west components of 24-hourly and 12-hourly periodic components varies in a systematic manner from season to season. In summer and autumn seasons the polar plots of the drift vector give highly elongated ellipses the major axes of which lie along east–west and north–south directions for the 24-hourly and

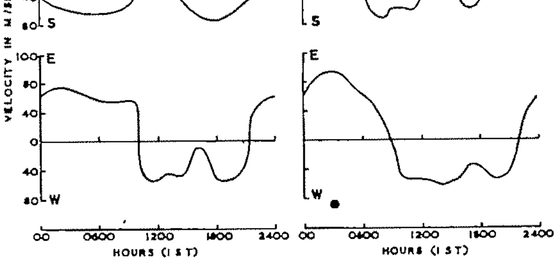

Fig. 1

12-hourly periodic components respectively. It can be seen from Table 1 that there is no radical change in the *E*-region drifts at Waltair for the two years.

Comparing these results with those obtained at some high-latitude stations, it may be noted that there are some points of disagreement. Unlike the results of drift measurements at high latitudes, where the 12-hourly component is found to be predominant, results from Waltair reveal a prominent 24-hourly component. The 24-hourly east–west component is directed towards east during a major part of the night and west during most of the daylight hours at this low-latitude station, while the behaviour is opposite for the high-latitude stations like Cambridge[1] and Kjeller[3]. The observation of an anti-clockwise sense of rotation of the 24-hourly drift vector for the northern hemisphere station is unusual and requires confirmation from data obtained at other low-latitude north and south hemisphere stations.

The 12-hourly north–south component shows maximum northward drift at 0300 and 1500 hr. at high-latitude stations while this occurs at 0000 and 1200 hr. local time at Waltair.

Recently, Skinner, Hope and Wright[4], while reporting results of drift measurements from Ibadan (geogr. lat. 07° 26′ N., geogr. long. 03° 53′ E.; geomag. 10·6° N.), have reproduced the curve of east–west component of *E*-region drifts during day-time, which shows features similar to our day-time curves for the east–west component. They have not observed any significant north–south component although their geomagnetic latitude is nearer to ours. The reason for the lack of north–south component at Ibadan is presumably due to the fact that the magnetic field at that station is nearly horizontal, the magnetic latitude being 2½° S.

A detailed paper embodying the results of this investigation will shortly be published elsewhere. We wish to acknowledge the financial assistance given by the Council of Scientific and Industrial Research for supporting these investigations.

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[1] Briggs, B. H., and Spencer, M., "Rep. Prog. Phys.", 17, 245 (1954).

[2] Rao, B. R., Rao, M. S., and Murty, D. S. N., *J. Sci. Indust. Res.*, A, 15, 75 (1956).

[3] Harang, L., and Pederson, K., *J. Geophys. Res.*, 62, 183 (1957)

[4] Skinner, N. J., Hope, J., and Wright, R. W., *Nature*, 182, 1363 (1958).

Life-Time of the Atomic Oxygen 6300 A. Line in the Auroral Spectrum

The red auroral lines of atomic oxygen at 6300 A. and 6364 A. are known to appear with a delay against other lines. The lag is due to the fact that the excited oxygen atoms are metastable, the life-time being about 110 sec.[1]. The delay of the oxygen 5577 line is for the same reason 0·75 sec.[2]. For experimental evidence records made by an automatic direct-recording spectrograph have been used and the variation of the red line due to changes in the luminosity of the aurora is compared with that of the N_2^+ band 4278 A. The green line 5577 A. may also be used; but on the spectral records this line was too often outside the record, because of over-loading of the amplifier.

Some examples of the records showing a sudden rise of the spectral intensity have been selected, and curves for both lines investigated are drawn with the same amplitude in order to measure the time difference at different moments. Fig. 1 shows three typical examples. The values of the delay are mean values received by measuring the time difference at a number of points for the rising part and the falling part of the curve respectively. Only examples with sufficiently close amplitude variations are used.

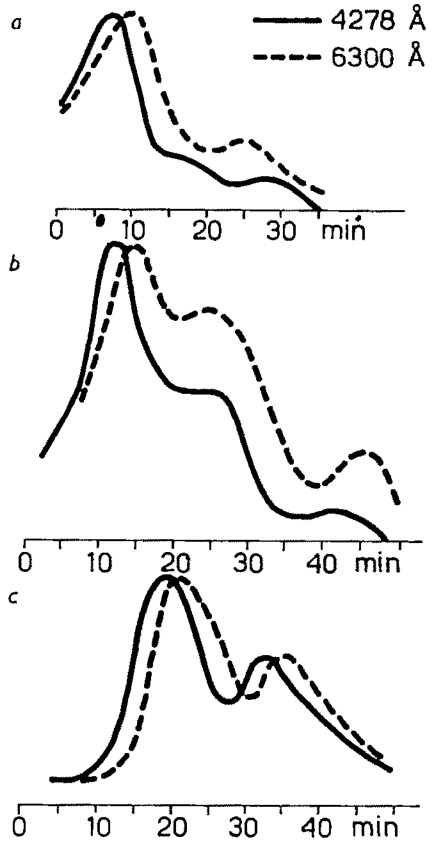

Fig. 1

The resulting mean value of 12 cases is $t_r = 110$ sec. for the rising part and $t_f = 175$ sec. for the falling part of the curve. These values may be regarded as approximate, for intensity variations of the aurora between each record and movements of the aurora will introduce some error. Further, the difference in height level of the excitation of both lines may influence the result. Nevertheless, it seems that the delay at the beginning of the emission of the red line is in accordance with the theoretical value but that the delay is greater when the emission decreases. The theoretical value of 110 sec. is a mean value of a large number of single processes and a slower fall of the curve is therefore quite reasonable.

Table 1

Curve No.	t_r	t
1	99	168
2	102	195
3	105	222
4	115	109
5	120	180
6	127	330
7	53	135
8	75	180
9	135	120
10	135	135
11	135	160
12	135	—
Mean values	$t_r = 110$ sec.	$t_f = 175$ sec.

Table 1 shows the spreading of values of the 12 cases analysed. In all, 36 cases have been investigated and the mean value of the measured delay including both the rising and falling period is ~120 sec.

We wish to thank Dr. A. Omholt for valuable discussion concerning this communication.

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[1] Garstang, R. J., *Mon. Not. Roy. Astro. Soc.*, **111**, 115 (1951).

[2] Omholt, A., and Harang, L., *J. Atmos. Terr. Phys.*, **7**, 247 (1955).

METALLURGY

Dependence on Temperature of Elastic Moduli of Tin

The crystalline structure of tin changes from cubic (grey tin) to body-centred tetragonal (white tin) at 18° C. These structural changes were examined as reflected in the thermoelastic behaviour of polycrystalline tin.

The composite piezo-electric oscillator method[1] has been employed in the present investigation, the frequency-range being 100–145 kc./sec. Specimen rods of rectangular and cylindrical cross-sections are cut from 99 per cent pure tin. The composite oscillator with its holder is placed at the centre of an electrical furnace and the temperature is controlled by varying the current using a variac. For study in the low-temperature region, the crystal holder sealed in a container is maintained in a Dewar flask containing freezing mixtures.

The observed variations of Young's modulus (Y) and rigidity modulus (G) with temperature are represented graphically in Fig. 1. The elastic moduli decrease linearly with temperature. A sudden change in the gradient of the curves occurs at about 18° C. in both the cases. This transition is due to the structural changes.

In addition to the above transition, the Young's modulus versus temperature curve shows another sudden change in the gradient at 80° C. It is quite possible that this phenomenon is due to the stress relaxation at grain boundaries. It is known that the temperature at which the grain boundary slipping takes place varies with the frequency of excitation.

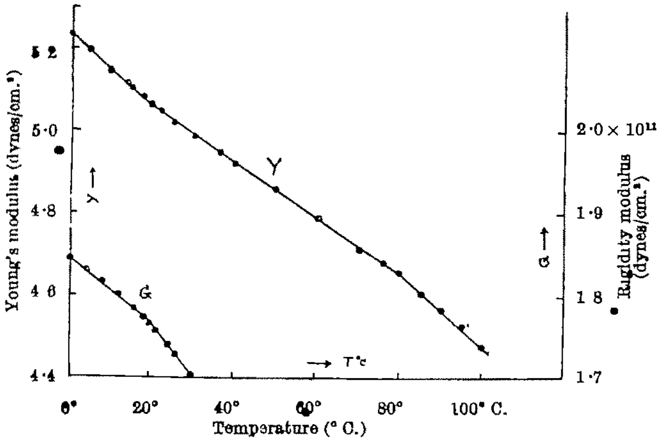

Fig. 1

Experimental investigations to study the thermoelastic behaviour of tin at low frequencies (3–10 kc./s.) are in progress. Such a study enables us to explain the mechanism responsible for the transition occurring at 80° C.

The volume compressibility of cubic tin at 0° C. is calculated making use of the values of the elastic moduli of polycrystalline tin at 0° C. The value works out to be $8{\cdot}31 \times 10^{-13}$ cm.^{2}/dyne.

We thank Dr. J. Bhimasenachar for his guidance and encouragement.

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[1] Subrahmanyam, S. V., *J. Chem. Phys.*, **22**, 1562 (1954).

Distortions in Metallic Structures

A puzzling feature of the Periodic Table has been that certain of the elements, such as indium, cadmium and zinc, while possessing typically metallic properties, have structures which are slight distortions of close-packed metallic structures.

Experimental[1] and theoretical[2] work on indium shows that the tetragonal distortions in this metal, and its alloys with various solutes, can be ascribed to a lowering of Fermi energy brought about by the distortion. The energies of the {002} overlaps for the face-centred cubic Brillouin zone are identical, and provided that the total number of electrons in these overlaps is below a certain value, this particular 'degeneracy' can be removed or lowered, with a lowering of Fermi energy, by orthorhombic or tetragonal distortions, at constant volume, of the metal lattice. The energetics of these distortions are similar to those presented by Dunitz and Orgel[3] for the Jahn-Teller distortions which have been so successful in explaining the structures of transition-metal oxides, though without the corresponding bonding and anti-bonding implications.

The energy scheme of Fig. 1*a* will enable this to be readily visualized; the levels shown are those corresponding to the initiation of the overlaps on (002) and (200) for a face-centred cubic structure, and for the tetragonally distorted structures with axial ratios respectively less than and greater than 1. The sequence of phases indicated by this scheme is observed in the systems indium–tin and indium–lead[4], in which the volume differences between the phases are very small.

It should be emphasized that in each structure the continuous energy band of the lower level actually overlaps that of the higher level; calculations (Wood, V. E., private communication) show that some states in the higher-energy overlap may be occupied without greatly affecting the stability conferred on the structure by the Fermi energy. The energy distribution curves in Fig. 1*b* are approximated by rectangles, so that the total energy of the electrons in these overlaps (proportional to the area below the Fermi surface) can be easily visualized. In this scheme, the first structure would become unstable when the band was filled to level A, and the stability of the cubic structure would equal that of the other two at level B.

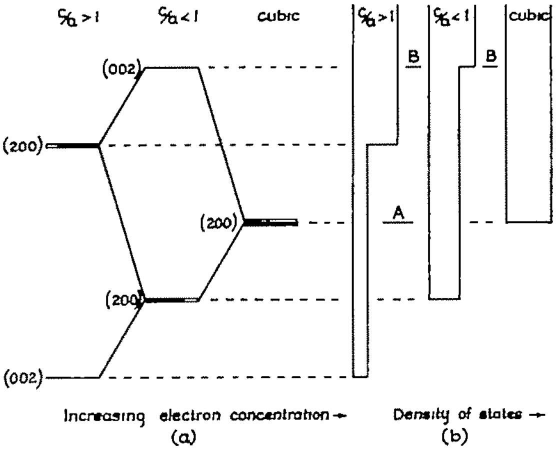

Fig. 1. Energy schemes for {002} Brillouin zone overlaps in metals with face-centred cubic and closely related tetragonal structures

A similar type of electronic stabilization may account for the large axial ratios of the close-packed hexagonal structures of zinc and cadmium, in which the low-multiplicity (0001) overlaps are moved to lower energies by the type of distortion observed.

Wood's calculations[2], based on electrostatic and Fermi energies only, give no indication of the factors which govern the equilibrium axial ratio in the cases considered here, and which prevent such distortions in the typical metallic structures. This may be due to the simplifying approximations made; but in my opinion, the origin of these balancing forces must be sought in the underlying assumptions.

Apart from an entropy effect which would be similar in all cases, there appear to be only two possibilities:

(*a*) In spite of the relatively open nature of the metals concerned, the properties of the ions themselves and/or their exchange interactions may be of importance. Some possible support for this view is given by the observation[5] that even the ion-cores in zinc seem to be distorted by elongation in the direction of the *c*-axis.

(*b*) A partial localization of the electron cloud may modify the full-zone contribution to the lattice energy. This would perhaps correspond to a change in energy due to the distortion of one type of bond in the resonant bond theory of metals.

In contrast to previous theories of the tetragonal structure of indium[6], the present theory easily explains the low-energy transformation to a second tetragonal phase which occurs in the systems indium–lead and indium–tin, and is thus to be preferred.

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[1] Graham, J., Ph.D. dissertation, University of Birmingham (1956). Tomasch, W. J., and Reitz, J. R., *Phys. Rev.*, 111, 757 (1958).

[2] Wood, V. E., U.S.A.E.C. Tech. Rep. No. 4, Case Institute of Technology, Contract *AT*(II-I)-623 (1958).

[3] Dunitz, J. D., and Orgel, L. E., *J. Phys. Chem. Solids*, 3, 20 (1957).

[4] Raynor, G. V., and Graham, J., *Research* (Lond.), 10, 369 (1957). Moore, A., Graham, J., Williamson, G. K., and Raynor, G. V., *Acta Met.*, 3, 579 (1955).

[5] McNatt, E. M., *Phys. Rev.*, 56, 406 (1939).

[6] Makarov, E. S., *Izvest Akad. Nauk S.S.S.R. Otdel Khim Nauk*, 485 (1950). Basinski, Z. S., and Christian, J. W., *Acta Met.*, 1, 754 (1953). Goodenough, J. B., and Loeb, A. L., *Phys. Rev.*, 98, 801 (1955).

CHEMISTRY

Electron Spin Resonance of γ-Irradiated, Substituted Toluenes

THE results reported here were obtained during an attempt to study the energy transfer efficiency from low-temperature, rigid glass solvent to dissolved solute molecules on γ-irradiation. This has recently been studied by Porter *et al.*[1], who recorded the ultra-violet absorption spectra of both γ-irradiated and ultra-violet illuminated side-chain substituted aromatic hydrocarbons dissolved in hydrocarbon glasses at 77° K. and, using comparative methods previously reported[2,3], assigned the various spectra to benzyl-type radicals. It was thought that using the electron spin resonance technique instead of ultra-violet spectroscopy, additional information might be obtained; but preliminary experiments soon showed that, while a radical signal characteristic of the solute could be detected, the fine structure of the electron spin resonance spectrum of the glass solvent made quantitative estimations impossible. However, the electron spin resonance spectra of the various radicals produced by γ-irradiation of the pure, degassed, frozen solutes alone were determined.

On first sight all the spectra seemed identical, being broad and featureless with negligible hyperfine splitting (Fig. 1). The signal was checked to be below saturation, and the narrow (about 10 gauss) super pure quartz reaction vessel signal—occasionally observed—was allowed for. However, on closer inspection it was found that the overall width differed significantly from radical to radical. Table 1 compares these results with those of Porter *et al.*[1].

With the exception of $C_6H_5CH_2CH_3$, which received additional irradiation, all the above were exposed and measured under as near as possible identical conditions, using the apparatus and techniques already described[4].

In order to obtain a very rough estimate of the *G*-value of the above radical production, 'cupric-ferrous' dosimetry was used[5] to establish that in 1 hr. the dose received was approximately 0 6 × 10^{18} × 100 eV./gm.; comparison of the signal from a known amount of diphenylpicrylhydrazyl in benzene with the above broad signals indicated that approximately 10^{18} spins/gm.—a lower limit depending on modulation and any (small) saturation effects—were formed after 1-hr. irradiation of all the samples except ethyl benzene and the quartz reaction vessel. The latter gave approximately 10^{15} spins/gm.

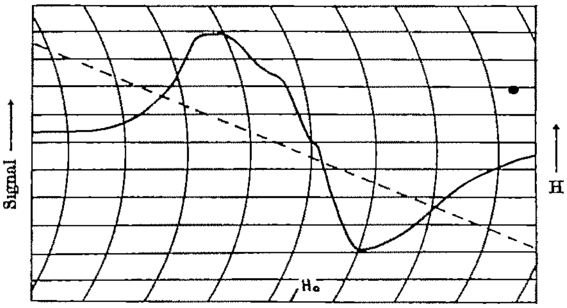

Fig 1. Electron spin resonance from isopropyl benzene after 20 min. γ-irradiation at 77° K. Spread, 34 gauss. Dashed line, magnetic field. Rate versus time

Table 1. ELECTRON SPIN RESONANCE OF SUBSTITUTED TOLUENES

Compound	Radical (see ref. 1) γ	Radical (see ref. 1) Ultra-violet	Electron spin resonance observed (and γ-irradiation time)
$C_6H_5.CH_3$	$C_6H_5.CH_2$	$C_6H_5.CH_2$	Broad, 46 gauss spread (1 hr.)
$C_6H_5.CH_2Cl$	$C_6H_5.CHCl$	$C_6H_5.CH_2$	,, 32 gauss (½ hr.) + 46 gauss (1 hr.)
$C_6H_5.CHCl_2$	$C_6H_5.CCl_2$	$C_6H_5.CHCl$	,, 66 gauss (1 hr.)
$C_6H_5.CCl_3$	$C_6H_5.CCl_2$	$C_6H_5.CCl_2$	,, 66 gauss (1 (hr)
$C_6H_5.CH_2CH_3$	$C_6H_5.CHCH_3$	$C_6H_5.CHCH_3$	,, (~80 gauss)—very weak (3 hr.)
$C_6H_5.C(CH_3)_3$	$C_6H_5.C(CH_3)_2$	$C_6H_5.C(CH_3)_2$	,, 32 gauss (20 min.)
$C_6H_5.CH(CH_3)_2$	$C_6H_5.C(CH_3)_2$	$C_6H_5.C(CH_3)_2$	,, 34 gauss (20 min.)

From the above minimum a value of $G \simeq 1{\cdot}6$ was obtained.

Inspection of Table 1 shows that there is a correlation between the electron spin resonance line spread in gauss and the radicals detected by Porter *et al.* by ultra-violet spectroscopy. In three cases in Table 1, Porter *et al.* found that two different compounds gave the same radical under suitable conditions. For these three cases the electron spin resonance of the pure γ-irradiated compounds has the same line-width. For benzyl chloride it can be seen that there are two species produced; this is also in agreement with the observations of Chilton and Porter[1] that a small amount of benzyl radical is formed in addition to the C_6H_5CHCl radical. However, though one can speculate as to the nature of the radicals responsible for the electron spin resonance, without fine structure no definite conclusions can be drawn.

I am greatly indebted to Dr. M. S. Matheson for helpful discussions, to Dr. B. Smaller for the loan of his apparatus and helpful discussions, to Mr. W. Mulac, who carried out the irradiations, and to Dr. E. Strachan for supplying purified samples.

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[1] Chilton, H. T. J., Porter, G., and Strachan, E. S., Farad. Soc. Informal Discussion on Free Radical Stabilization, Sheffield (1958). Chilton, H. T. J., and Porter, G., *J. Phys. Chem.*, **63**, 904 (1959).

[2] Norman, I., and Porter, G., *Proc. Roy. Soc.*, A, **230**, 399 (1955).

[3] Porter, G., and Strachan, E. S., *Trans. Farad. Soc.*, **54**, 1595 (1958).

[4] Smaller, B., and Matheson, M. S., *J. Chem. Phys.*, **28**, 1169 (1958).

[5] Hart, E., *Radiation Res.*, **2**, 33 (1955).

Phosphides of the Platinum Metals

THE existence of several phosphides of the platinum metals has been reported by Biltz *et al.*[1]. The knowledge of the physical and chemical properties of these phases is, however, very meagre. Crystal structures have been determined for only three phosphides, namely, Rh_2P and Ir_2P with the anti-fluorite structure[2] and PtP_2 with the pyrite structure[3].

I have started an X-ray powder and single-crystal investigation of phosphides of the platinum metals in order to establish the compositions of the phases and to determine the crystal structures. A preliminary report of the work is given below.

The phosphides have been prepared by heating metal powder (purity 99·5 per cent or higher) and red phosphorus in sealed and evacuated silica tubes at temperatures between 500° and 1,100° C. No appreciable attack on the silica tubes has been observed. Powder photographs of the reaction products have been taken with Guinier-type focusing cameras in copper $K\alpha$ radiation. Calcium fluoride ($a = 5{\cdot}4630$ A.) has been used as internal calibration standard on each powder photograph, and the accuracy of the lattice parameter determinations is estimated to be higher than 0·05 per cent.

From a survey of the X-ray powder data, the unit cell dimensions and crystal structure types of the phases Ru_2P, RuP, RuP_2, OsP_2, RhP_3, PdP_3 and IrP_3 have been determined. The earlier structure determinations of Rh_2P, Ir_2P and PtP_2 have been confirmed, and the unit cell dimensions have been redetermined. Crystallographic data for the above-mentioned phases are given in Table 1.

Table 1. CRYSTALLOGRAPHIC DATA OF SOME PLATINUM METAL PHOSPHIDES

Phosphide	Structure type	Space group	*a*	*b*	*c*
Ru_2P	*C*23 (anti-$PbCl_2$)	*Pnma* (*Pmnb*	5·902 3·859	3·859 5·902	6·896 6·896)
RuP	*B*31 (MnP)	*Pnma* (*Pbnm*	5·520 6·120	3·168 5·520	6·120 3·168)
RuP_2	*C*18 (marcasite)	*Pnnm* (*Pmnn*	5·115 2·870	5·888 5·115	2·870 5·888)
OsP_2	*C*18 (marcasite)	*Pnnm* (*Pmnn*	5·098 2·918	5·898 5·098	2·918 5·898)
Rh_2P	*C*1 (anti-fluorite)	*Fm*3*m*	5·498	—	—
Ir_2P	*C*1 (anti-fluorite)	*Fm*3*m*	5·543	—	—
RhP_3	*D*2 (skutterudite)	*Im*3	7·996	—	—
PdP_3	*D*2 (skutterudite)	*Im*3	7·705	—	—
IrP_3	*D*2 (skutterudite)	*Im*3	8·015	—	—
PtP_2	*C*2 (pyrite)	*Pa*3	5·694	—	—

The standard setting of the unit cells in "International Tables for X-ray Crystallography", 1 (1952) has been adopted. The settings of the unit cells according to the descriptions of the *C*23, *B*31 and *C*18 types in "Strukturbericht" are given in parenthesis. The unit cell dimensions are given in angstrom units.

The structural similarities with the phosphides of the iron group metals may be noted. Co_2P[4] is *iso*structural with Ru_2P, FeP[5] and CoP[5] are *iso*structural with RuP, FeP_2[6] is *iso*structural with RuP_2 and OsP_2, CoP_3[7] and NiP_3[7] are *iso*structural with RhP_3, PdP_3 and IrP_3. It may further be mentioned that the powder photographs of RhP_2 and IrP_2 show a very close resemblance, and it is thus highly probable that RhP_2 and IrP_2 are *iso*structural.

The crystal structure investigations of the remaining phosphides of the platinum metals will mainly be performed with single-crystal methods. Detailed reports of this work will be published later (*Acta Chem. Scand.*).

This investigation was made during the tenure of a generous scholarship from the Statens Tekniska Forskningsråd. The work has been supported in part by the Air Force Office of Scientific Research of the Air Research and Development Command, United States Air Force, through its European Office under contract No. *AF* 61(052)-40. I wish to thank Prof. G. Hagg for his kind interest.

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[1] Biltz, W., Weibke, F., May, E., and Meisel, K., *Z. anorg. Chem.*, **223**, 129 (1935). Wiehage, G., Weibke, F., Biltz, W., Meisel, K., and Wiechmann, F., *ibid.*, **228**, 357 (1936). Biltz, W., Ehrhorn, H.-J., and Meisel, K., *ibid.*, **240**, 117 (1939). Söffge, K. H., Helmbrecht, M., and Biltz, W., *ibid.*, **243**, 297 (1940). Faller, F.-E., Strotzer, E. F., and Biltz, W., *ibid.*, **244**, 317 (1940).

[2] Zumbusch, M., *Z. anorg. Chem.*, **243**, 322 (1940).

[3] Thomassen, L., *Z. physik. Chem.*, B, **4**, 277 (1929).

[4] Nowotny, H., *Z. anorg. Chem.*, **254**, 31 (1947).

[5] Fylking, K.-E., *Arkiv Kemi Min. Geol.*, B, **11**, No. 48 (1935).

[6] Meisel, K., *Z. anorg. Chem.*, **218**, 360 (1934).

[7] Rundqvist, S., and Larsson, E., *Acta Chem. Scand.*, **13**, 551 (1959).

Zone of Negative Temperature Coefficient in Hydrocarbon Oxidation

THE occurrence of a zone of negative temperature coefficient is a feature almost unique to hydrocarbon oxidation. It is most intriguing how the rate and extent of reaction, which at first increase rapidly with temperature and give rise to cool flames, begin to fall with further rise in temperature. Although, justifiably, there has been a great deal of speculation about the factors responsible for this zone, very little experimental work seems to have been done to explore it.

We have recently investigated the zone of negative temperature coefficient of a number of hydrocarbons. The effect of various additives, which represent different categories of reaction intermediates, such as peroxides, aldehydes, ketones, alcohols and olefins, has also been examined. The experimental conditions used were similar to those described previously[1].

In general, the zone of negative temperature coefficient has been found to extend over the same temperature range—approximately 380–460° C.—for different hydrocarbons. There are, however, marked differences with respect to the magnitude of the coefficient. Among *n*-paraffins (pentane to dodecane) the magnitude of the negative temperature coefficient decreases with increasing chain-length, the effect being particularly marked among the lower members of the series. In branched paraffins (for example, isomeric hexanes) the magnitude of the coefficient tends to increase with the degree of branching. Furthermore, the temperature-range between the onset of the negative temperature coefficient and the commencement of the following region of positive temperature coefficient also increases.

The addition of di-*t*-butyl peroxide has a pronounced effect on the zone of negative temperature coefficient. It markedly enhances the extent of reaction and lowers the magnitude of the coefficient. It also reduces the temperature-range over which the region extends. Acetaldehyde and propionaldehyde also affect the zone of negative temperature coefficient in the same manner as di-*t*-butyl peroxide, although the effect of both aldehydes is considerably less than that of the peroxide. Acetaldehyde is more effective than propionaldehyde. The addition of acetone has scarcely any effect, while methyl ethyl ketone slightly enhances the extent of reaction. Methanol and ethanol have, if anything, a slightly inhibitory effect on the reactions.

Different olefins markedly affect the zone of negative temperature coefficient: they may act either as inhibitors or accelerators. The olefins which inhibit reactions (such as ethylene, *iso*butene and 2,4,4-trimethylpentene-1) can be recognized as those resistant to oxidative degradation in the temperature-range of the negative temperature coefficient, and furthermore their saturated analogues are also unreactive in this region (Salooja, K. C., unpublished work). On the other hand, the olefins which accelerate reactions (such as butene-1, butene-2 (*cis* or *trans*), hexene-1 and hexa-1,5-diene) are those which by themselves undergo oxidation, and/or the saturated analogues of which are reactive in this region (Salooja, K. C., unpublished work).

It is clear from the effect of various types of additives that olefins of the first category mentioned above are capable of causing marked inhibition of the reactions. This suggests that such olefins may themselves be responsible for causing the onset of the negative temperature coefficient. That such olefins are indeed formed in appreciable amounts in the cool flame region preceding the zone of negative temperature coefficient is well known[2-4]. Moreover, it has also been shown that the concentration of olefins increases with temperature[5,6].

The formation of olefins in the temperature-range of the zone of negative temperature coefficient is expected from (1) the low stability of RO_2 radicals beyond the cool flame region, $RO\cdot_2 \rightarrow$ olefin + $HO\cdot_2$, and (2) a split in the carbon–carbon bond of the alkyl radical, $R\cdot \rightarrow$ olefin + $R'\cdot$. Apart from cases where the RO_2 radical corresponds to a lower carbon number fragment of the starting material, it is particularly the olefin generated by the second reaction that would be responsible for the onset of the negative temperature coefficient. This is so because only the olefins of lower carbon number (and also their saturated analogues) are likely to be more resistant to oxidation than the parent hydrocarbon.

Although analytical data are not yet available on the combustion products of all the hydrocarbons investigated so far, it does not seem unreasonable to suggest that the observed relative order of negative temperature coefficients of different hydrocarbons could be related to the type of olefins that could arise predominantly from different hydrocarbons. For example, among *n*-paraffins the lower members of the series would no doubt lead to the formation of lower carbon number olefins than the higher members of the series. Since the lower olefins, and also their saturated analogues, are more resistant to oxidation than the higher ones (ref. 7 and Salooja, K. C., unpublished work), the lower *n*-paraffins, as observed, would have a negative temperature coefficient of higher magnitude. Again, in comparison with *n*-paraffins, the branched-chain paraffins would lead to the formation of branched olefins (which are more resistant to oxidation than the corresponding straight-chain olefins (Salooja, K. C., unpublished work), and the saturated analogues of which are also far more resistant to oxidation than those of the corresponding straight-chain olefins[4,7]) and hence, in accordance with the observations, the branched-chain hydrocarbons would have a sharper negative temperature coefficient.

The effect of various additives can be readily interpreted by considering it *vis-à-vis* the chain-terminating effect of the olefins that inhibit reactions. Additives that are unreactive in the temperature range of the negative temperature coefficient would have no effect in this region. Reactive additives like acetaldehyde and, even more so, di-*t*-butyl peroxide, which can markedly increase chain-branching species, would, as observed, lower the magnitude of the negative temperature coefficient.

Apart from the role of olefins, it is possible, as suggested by several investigators (notably Satterfield and Wilson[5]), that the expected decrease in the number of highly reactive OH radicals relative to HO$_2$ radicals at temperatures above the cool flame region may also contribute to the onset of the zone of negative temperature coefficient. Further work is in progress and the results will be reported in detail elsewhere.

I wish to thank the Directors of 'Shell' Research, Ltd., for permission to publish this work.

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[1] Salooja, K. C., *Nature*, 183, 602 (1959).

[2] Bailey, H. C., and Norrish, R. G. W., *Proc. Roy. Soc.*, A, 212, 311 (1952).

[3] Neu, J. T , *J. Phys. Chem.*, 60, 320 (1956)

[4] Boord, C. E., "Literature of the Combustion of Petroleum", Advances in Chemistry Series, No. 20, 5 (Amer. Chem. Soc., Washington, 1958).

[5] Satterfield, C. N., and Wilson, R. E., *Indust. Eng. Chem.*, 46, 1001 (1954).

[6] Frank, C. E., and Blackham, A. U., *Indust. Eng. Chem.*, 46, 212 (1954).

[7] Cullis, C. F., and Hinshelwood, C. N., *Disc. Farad. Soc.*, 2, 117 (1947).

BIOPHYSICS

Aggregation and Staining Behaviour of Ultra-violet Irradiated Films of Nucleic Acids

During a study of the staining behaviour of ultra-violet irradiated films of deoxyribonucleic acid (deposited by evaporation from aqueous solution on glass slides and exposed to the 2357 A. emission of a 30-watt germicidal lamp), it was found that, upon exposure of an irradiated film to water or salt solution, the deoxyribonucleic acid passes into the aqueous phase as insoluble fibres readily visible to the naked eye. For a film containing 25 µgm. deoxyribonucleic acid spread over an area 10 mm. in diameter, and with an incident intensity of 10^4 ergs/cm.^{2}/sec., fibre formation is visible following several seconds irradiation. With increasing doses the dimensions of the fibres increase until, after 1–2 hr., they are 7–8 mm. in length and 200µ in diameter; beyond this point further irradiation results in degradation.

Fibre formation may also be observed on the glass slides directly under the microscope or, better still, after staining with a basic dye. Analogous results are obtained with highly polymerized deoxyribonucleic acid from widely different sources.

In some respects this phenomenon resembles that reported for commercial deoxyribonucleic acid films irradiated *in vacuo* or under dry nitrogen with the subsequent formation, in water or salt solution, of a gel susceptible to the action of deoxyribonuclease[1]. In the experiments reported here the fibres were completely insoluble in water or heated salt solution and were completely resistant to the prolonged action of deoxyribonuclease.

The apurinic acid resulting from the mild acid hydrolysis of deoxyribonucleic acid has a molecular weight of only 15,000[2] and hence a chain-length about 1 per cent that of the parent deoxyribonucleic acid. But irradiated apurinic acid forms fibres similar in magnitude to those obtained with deoxyribonucleic acid. The ultra-violet induced aggregation (polymerization) or cross-linking is consequently not dependent on the use of highly polymerized material, but involves the formation of specific intermolecular linkages. The importance of the pyrimidine rings in this reaction is testified to by the fact that polyadenylic and polyguanylic acids do not exhibit fibre formation whereas polyuridylic acid does. Ribonucleic acid reacts to only a slight degree and proteins and polysaccharides not at all.

The staining behaviour of these irradiated films is of considerable interest, particularly with regard to methyl green. Dye uptake and retention were measured quantitatively by use of dyes labelled with carbon-14 and sulphur-35, followed by end-window counting[3]. It was found that the affinity of deoxyribonucleic acid for methyl green increases by as much as 50–80 per cent upon irradiation, following which it decreases. Particularly striking is the behaviour of apurinic acid, the affinity of which for methyl green is very low; a film of apurinic acid irradiated under exactly the same conditions as a deoxyribonucleic acid film will bind as much, or even more, dye than the latter. Staining with other dyes showed that increased binding of methyl green is not due to trapping of dye molecules in the fibre interstices.

Deoxyribonucleic acid films stain Gram-negative, as compared to apurinic acid and ribonucleic acid, which are Gram-positive[4]. However, an irradiated deoxyribonucleic acid film is so strongly Gram-positive that its decolorization time approximates that prevailing for numerous Gram-positive micro-organisms.

The above, as well as other, results, which will be fully reported elsewhere, are of interest from several points of view.

(*a*) We have here an example of radiation-induced aggregation (cross-linking) of a biological polymer by ultra-violet light like that already observed for such synthetic polymers as poly(vinylchloride)[5], polyethylene and polyacrylamide[6]. In preliminary experiments we have found that ionizing radiations (γ-rays from cobalt-60 source) do not promote aggregation in either deoxyribonucleic acid or apurinic acid (cf., however, ref. 1).

(*b*) A nucleic acid film in air is, in some respects, more representative of the state of deoxyribonucleic acid in living cells than is a solution of deoxyribonucleic acid. We have indeed found some correlation in staining behaviour between irradiated nucleic acid films and some micro-organisms and tissue sections. It would obviously be desirable to examine the irradiated deoxyribonucleic acid, apurinic acid and polyuridylic acid films by X-ray diffraction.

(*c*) The extent to which deoxyribonucleic acid binds methyl green, which is of considerable importance in cytochemistry, is seen to be dependent not only on the molecular weight of the nucleic acid but also on its state of aggregation which, in cell nuclei, may be quite high. Irradiated films of deoxyribonucleic acid should also be useful for *in vitro* studies of the mechanism of Gram staining[4].

We are indebted to Dr. S. Ochoa and Dr. R. F. Beers for gifts of enzymatically synthesized polyadenylic acid and to Dr. A. M. Michelson for chemically synthesized polyuridylic and polyguanylic acids.

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[1] Setlow, R., and Doyle, B., *Biochim. Biophys. Acta*, **15**, 117 (1954).

[2] Tamm, C., and Chargaff, E., *J. Biol. Chem.*, **203**, 689 (1953).

[3] Shugar, D., Proc. Second Inter. Conf. Peaceful Uses of Atomic Energy, Geneva, **24**, 263 (1958).

[4] Shugar, D., and Baranowska, J., *Biochim. Biophys. Acta*, **23**, 227 (1957); Inter. Conf. on Radioisotopes in Scientific Research, Unesco, Paris, 1957, Paper No. 149 (Pergamon Press, London, 1958).

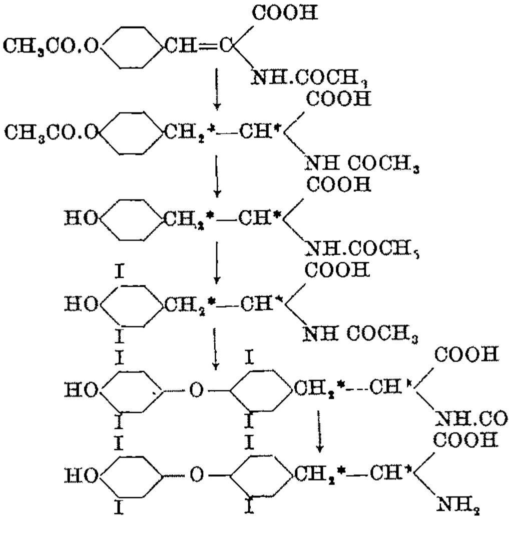
[5] Sobue, H., Tabata, Y., and Tajima, Y., *J. Polymer Sci.*, **27**, 596 (1958).

[6] Oster, G., Oster, G. K., and Moroson, H., *J. Polymer Sci.*, **26**, 233 (1957).

BIOCHEMISTRY

Synthesis of Tritium-labelled Tyrosine, 3 : 5-Diiodotyrosine and Thyroxine

ALTHOUGH much of the progress in thyroid hormone biochemistry is due to the use of thyroxine and 3 : 5 : 3′-triiodothyronine labelled with iodine-131, the high metabolic lability of the iodine atoms makes it impossible to follow the fate of these substances beyond the stage of enzymic deiodination[1]. The low specific activity of thyroxine labelled with carbon-14[2] has ruled out its use under physiological conditions. Hence, with the availability of pure tritium gas and methods for its measurement, it was decided to prepare tritium-labelled thyroxine. The commonly used methods of exchange could not be used because of the danger of reductive deiodination by tritium gas. The procedure summarized below, and which involved the synthesis of N-acetyl-tyrosine and N-acetyl-3 : 5-diiodotyrosine as intermediates, was finally adopted. All compounds were hence labelled in 2 carbon atoms of their alanine side-chain.



4-Acetoxy-α-acetamidocinnamic acid, synthesized according to Saul and Trikojus[3], was dissolved in dioxan and reduced with tritium gas in the presence of platinum oxide to O,N-diacetyl-tyrosine. (This reaction was carried out by Dr. E. Anthony Evans at the Radiochemical Centre, Amersham.) The specific activity of O,N-diacetyl-tyrosine was 178·8 mc./m.mole. Tritium-labelled tyrosine could be obtained by its hydrolysis.

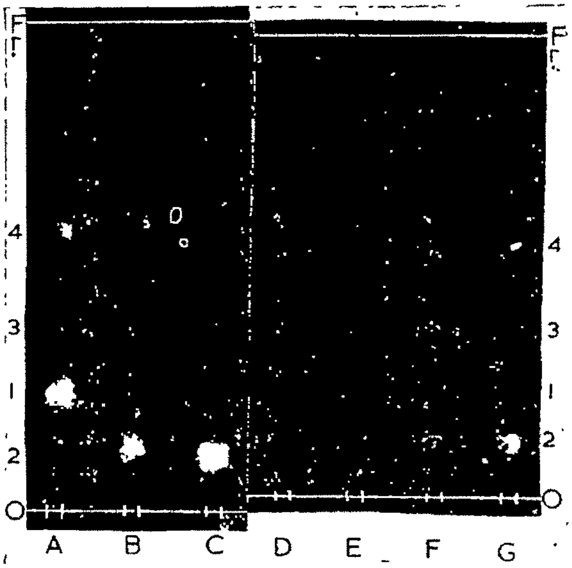


Fig. 1 Autoradiogram of a chromatogram developed to illustrate the reactions leading to the formation of ^{3}H-labelled N-acetyl-thyroxine. *A*: N-acetyl tyrosine in ammoniacal solution; *B*: After the addition of 1·5 m.mole iodine to the ammoniacal solution; *C*: after the addition of 5·0 m.mole iodine; *D*: 24 hr. after the incubation at 37° of reaction products in solution *C*, *E*: 4 days of incubation; *F*: 8 days; *G*: 12 days; 1: N-acetyl-tyrosine; 2: N-acetyl-3 : 5-diiodotyrosine; 3: N-acetyl-thyroxine; 4: unknown tritium-labelled material present as contamination in the original sample of O,N-diacetyl-tyrosine. Chromatographic solvent: *n*-butanol (80)-dioxan (20)-2*N* ammonia, *O*: origin of chromatogram; *Fr*: solvent front. Small amounts of radioactive products formed as a result of self-radiation damage are not visible in the autoradiogram

For the synthesis of labelled diiodotyrosine and thyroxine, 0·150 m.mole O,N-diacetyl-tyrosine was partially hydrolysed in 0·325 ml. 1 *N* sodium hydroxide at room temperature for 30 min. to N-acetyl-tyrosine. 0·350 ml. of concentrated ammonia (sp. gr. = 0·880) solution was added after neutralization of sodium hydroxide with 0·110 ml. 3·5 *N* hydrochloric acid. To this was added dropwise 0·5 m.mole iodine in potassium iodide at 0° and the excess iodine was neutralized with $Na_2S_2O_5$. The N-acetyl-3 : 5-diiodotyrosine formed is separated by preparative paper chromatography if diiodotyrosine is to be obtained. It is not separated if thyroxine is to be prepared; instead the volume of the solution was reduced to 0·3 ml. to which 1 ml. of 0·05 *M* borate buffer, *p*H 7·9, was added. Incubation of this solution at 37° C. led to the coupling of 2 molecules of N-acetyl-diiodotyrosine to 1 molecule of N-acetyl thyroxine, as described by Pitt-Rivers[4]. At the end of 12 days of incubation, the tritiated N-acetyl-thyroxine, the uncoupled N-acetyl-diiodotyrosine and other radioactive products were separated by preparative paper chromatography. Paper chromatography and autoradiography of aliquots withdrawn at different stages of the synthesis were used to follow the course of reaction as shown in Fig. 1.

Autoradiography was found to be a most convenient method for the detection and estimation of tritiated substances on chromatograms. Liquid scintillation counting confirmed the validity of such an estimation. The yield of tritiated N-acetyl-thyroxine was low: 0·009 m.mole (specific activity 56 mc./m.mole). Half the radioactivity would be lost in one alanine side-chain split off in the condensation of two molecules of N-acetyl-diiodotyrosine. The N-acetyl group was hydrolysed with 5 N hydrochloric acid in the presence of iodide to prevent deiodination. Only the racemic forms of thyroxine, diiodotyrosine and tyrosine could be prepared by the above method. All attempts to obtain the L-forms by the action of L-amino-acid de-acetylase from pig kidney or penicillium extracts have so far failed.

We are indebted to Dr. R. Pitt-Rivers for help and guidance.

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[1] Pitt-Rivers, R., and Tata, J. R., "The Thyroid Hormones" (Pergamon Press, London, 1959)

[2] Lipner, H. J., Barker, S. B., and Winnick, T , *Endocrinol* , 51, 406 (1952).

[3] Saul, J. A., and Trikojus, V. M., *Biochem. J.*, 42, 80 (1948).

[4] Pitt-Rivers, R , *Biochem. J.*, 43, 223 (1948) Pitt-Rivers, R., and James, A. T., *ibid.*, 70, 173 (1958).

Inhibition of Lipid Peroxidation in Microsomes by Vitamin E

The requirement for vitamin E is largely represented by a need for a biologically active lipid antioxidant. Vitamin E-deficient animals show *in vivo* lipid peroxidation and a wide variety of metabolic changes[1]. How metabolic changes are related to lipid peroxidation is a major problem.

Recently, we found[2] that isolated mitochondria deteriorate by hæmatin-catalysed lipid peroxidation. In addition, mitochondria isolated from the livers of vitamin E-deficient rabbits had undergone lipid peroxidation *in vivo* as measured by the thiobarbituric acid reaction. When isolated these mitochondria were more labile to lipid peroxidation *in vitro* than those from controls. This suggests that the free-radical intermediates of lipid peroxidation damage the mitochondria structurally and functionally, thus bringing about deranged metabolism.

Microsomes should be labile to lipid peroxidation for the same reasons as mitochondria. The microsomal fraction contains 30–40 per cent total lipid, mainly phospholipides, and contains twice the amount of polyunsaturated lipids on a nitrogen basis as do mitochondria. Some of these lipids are in close molecular proximity to cytochrome b_5. Cytochromes are among the most potent lipid peroxidation catalysts. Vitamin E is the only known lipid antioxidant in microsomes where it would function to inhibit lipid peroxidation.

Following the same course of research with microsomes as with mitochondria[2] it was found that the microsome fraction from rat livers deteriorated by lipid peroxidation at a rate of 0·1 ml. oxygen reacted/mgm. nitrogen/hr. at 37° C. Increase in thiobarbituric acid reactants paralleled oxygen absorption during the first 2 hr. Added α-tocopherol at 0·25 mgm./mgm. nitrogen gave 72 per cent inhibition of oxygen absorption and 69 per cent inhibition of thiobarbituric acid reactants. Since 0·01 M cyanide and 0·1 M methylene blue give 71–94 per cent inhibition as measured by oxygen absorption or thiobarbituric acid reactants, the lipid peroxidation appears to be catalysed by cytochrome b_5 and other hæmochromes present. Cytochrome b_5 destruction in a 4-hr. reaction was 80 per cent. To determine if the microsomes of vitamin E-deficient animals undergo lipid peroxidation *in vivo* they were isolated from the livers of vitamin E-deficient and control rabbits. The packed volume yield was one-third and the total nitrogen/ml. was six times higher for the microsomes from the vitamin E-deficient as compared to controls. The significance of these differences is at present unknown. Microsomes from vitamin E-deficient rabbits had 5·7 times as much thiobarbituric acid reactants per ml. as the controls. This highly significant difference ($P = 0·001$) indicates that the microsomes had undergone lipid peroxidation *in vivo*.

Lipid peroxidation damage to the microsomes of vitamin E-deficient animals may account for degeneration of microsomes as observed by electron microscopy[3], increased turn-over rate of some nucleic acids[4], effects on protein synthesis[1], and decreased ascorbic acid synthesis[5].

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[1] Harris, P. L., and Mason, K E., in "Vitamin E", 3rd International Congress on Vitamin E, Venice, Italy (1955).

[2] Tappel, A. L., and Zalkin, H., *Arch Biochem. Biophys* , 80, 326, 333 (1959)

[3] Piccardo, M. G., and Schwarz, K., in "Liver Function, a Symposium on Approaches to the Quantitative Description of Liver Function" (Amer. Inst. Biological Sci., Washington, D.C., 1958).

[4] Dinning, J. S., *J. Biol. Chem.*, 212, 735 (1955).

[5] McCay, P. B., Carpenter, M. P., Kitabchi, A. E., and Caputto, R., *Arch. Biochem. Biophys.*, 82, 472 (1959).

Biosynthesis of Glycerides in the Mucosa of the Small Intestine

It is generally agreed that a mixture of free fatty acids, glycerol, monoglycerides and diglycerides is absorbed by the intestinal cells during digestion of fat and that the chyle secreted by these cells contains mainly triglycerides[1]. The present communication describes some studies on the biosynthesis of triglycerides by mitochondrial preparations from the mucosa of the small intestine of the rabbit and the rat.

For the preparation of the mitochondria, the small intestine was removed and washed with ice-cold potassium chloride (1·3 per cent). The mucosa was scraped off, homogenized in 10–15 vol. of ice-cold sucrose (0·25 M), and the mitochondrial fraction obtained by the usual sedimentation methods[2]. The washed mitochondria were suspended in potassium phosphate buffer (0·05 M, pH 7·0). 1·0 ml. of this suspension (containing between 1 and 4 mgm. protein per ml.) was incubated for 1 hr. at 37° C. with palmitic acid labelled with carbon-14 and various co-factors (see Table 1). Pure DL-α-glycerophosphate and labelled palmitic acid were used in aqueous solution as their potassium salts. α-Monostearin and α,β-dipalmitin were emulsified with small amounts of potassium oleate (0·8 μmole per 16 μmoles of α-monostearin; 1·2 μmoles per 4·0 μmoles of α,β-dipalmitin). All co-factors were at pH 7·0.

Reaction was stopped by the addition of 1/10 vol. of 50 per cent trichloracetic acid and the precipitate washed with water. The lipids were extracted with organic solvents and purified on alumina columns. The glyceride fractions were taken to dryness and counted as described elsewhere[3].

Experiments 1 and 2 show that α-monostearin, α,β-dipalmitin and α-glycerophosphate all stimulated the incorporation of labelled fatty acid into glycerides

Table 1

Exp.	Additions (μmoles)	Activity (disintegrations/ min./test)
1	None*	1,740
	α-monostearin (16)†	1,750
	α-monostearin (16) + all co-factors‡	2,515
	α-glycerophosphate (7)†	1,760
	α-glycerophosphate (7) + all co-factors	8,100
2	α,β-dipalmitin (4)†	1,600
	α,β-dipalmitin (4) + all co-factors	2,120
3	α-glycerophosphate (7)†	1,270
	α-glycerophosphate (7) + all co-factors	3,760
	,, omit only GSH	3,850
	,, omit only $MgCl_2$	2,180
	,, omit only CoA	1,760
	,, omit only ATP	1,350

Each system contained: 1·0 ml. of rabbit mitochondrial suspension in 0·05 *M* potassium phosphate buffer, *p*H 7 0, and 0 6 μM of labelled palmitic acid (0 4 μc.). After the additions described above, the final volume was adjusted to 3·0 ml with distilled water. Incubation time was 60 min. To those systems which did not contain monoglyceride initially, 16 μM of α-monostearin in aqueous solution (0 4 ml.) was added immediately prior to precipitation with trichloracetic acid. This was to act as a carrier for any radioactive lipid formed.

* Reaction stopped after zero minutes of incubation. This represented the level of unchanged labelled palmitic acid coming through the isolation procedure. This figure varied with each separate experiment, and was approximately 0·1 per cent of the original fatty acid added.

† This value was taken as the blank for the experiment, and was almost identical to the activity of the zero time system.

‡ 'All co-factors' refers to the following additions: ATP (5), CoA (0 25), GSH (10), $MgCl_2$ (10).

Abbreviations: ATP, adenosine triphosphate; CoA, coenzyme *A*; GSH, reduced glutathione.

when supplemented with adenosine triphosphate, coenzyme *A*, reduced glutathione, and magnesium chloride. α-Glycerophosphate gave an apparently greater stimulation than the lower glycerides. This could partially be explained by a dilution effect if the potassium oleate (added to emulsify the glycerides) was incorporated in significant amounts. If the rate for this was the same as that for labelled palmitate, then the relative rate for α-glycerophosphate to α-monostearin (Exp. 1) would be 100 to 30. In experiments 2 and 3 (the same mitochondrial suspension was used in both) the relative rate for α-glycerophosphate to α,β-dipalmitin would be 100 to 64. These ratios do not take into account the specific activities of the radioactive glycerides synthesized from these precursors. Since the specific activity of the triglyceride formed from α-glycerophosphate is three times that from α,β-diglyceride, the relative rate for these becomes 100 to 192. Furthermore, the lower glycerides might be less available than glycerophosphate since they were added as emulsions.

When a mixture of α-monostearin and α-glycerophosphate was added to the system, the incorporation was found to be the same as the sum of that given by either separately.

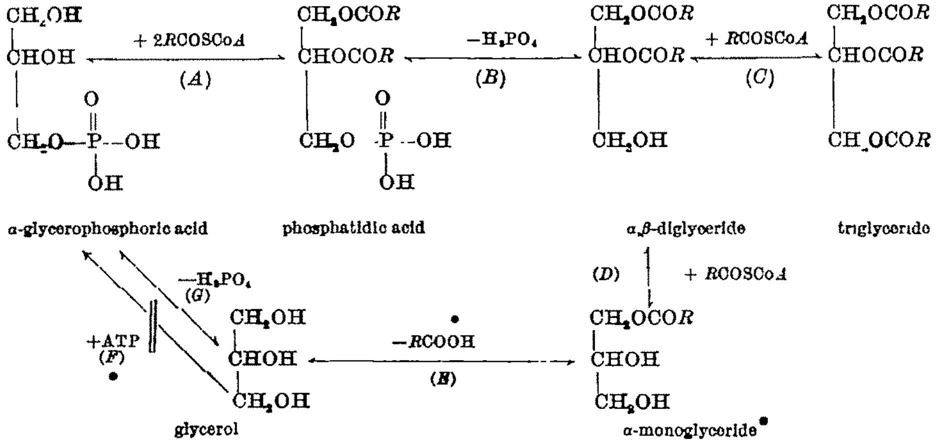
Experiment 3 shows the need for certain co-factors; there is a definite requirement for adenosine triphosphate, coenzyme *A* and magnesium chloride, but not for glutathione. It is possible that the mitochondria can reduce the coenzyme *A* which had not been reduced before addition.

The radioactive glycerides formed from α-glycerophosphate were separated on silicic acid columns. 90 per cent of the radioactive lipids were eluted from the columns in the positions where tri- and di-glycerides have been shown to come.

The above results indicate a pathway similar to that suggested by Stein, Tietz and Shapiro[4] for the synthesis of triglycerides in rat liver mitochondria, where α-glycerophosphate and phosphatidic acid are precursor and intermediate respectively. Dawson and Isselbacher described in a report of preliminary experiments[5] the incorporation of fatty acids labelled with carbon-14 into neutral fat using homogenates of the mucosa of the rat small intestine. They found that the incorporation was stimulated by adenosine triphosphate, coenzyme *A* and magnesium ions. Buell and Reiser[6] showed that α-glycerophosphate was involved in the biosynthesis of glycerides in subcellular fractions of the swine small intestine.

The appreciable incorporation found with monoglyceride would suggest an additional pathway, that is, the direct esterification of monoglyceride to diglyceride (reaction *D*). Though the monoglyceride might be hydrolysed (*E*), the phosphorylation of glycerol to α-glycerophosphate by adenosine triphosphate (*F*) does not seem to occur in intestinal extracts[6]. Adenosine triphosphate, coenzyme *A* and magnesium ions are required for the formation of activated long-chain fatty acids which participate in reactions *A*, *C* and *D*. In the present work, the fatty-acyl–coenzyme *A* synthetase was demonstrated in mitochondrial preparations of intestinal mucosa using the assay system described by Kornberg and Pricer[7].

Similar studies were carried out using mitochondrial preparations from rat intestine; but here it was found that α-glycerophosphate did not stimulate the incorporation of labelled palmitate to any great extent. The relative rates obtained were: α-monoglyceride to α-glycerophosphate, 100 to 10, giving further evidence for the occurrence of reaction *D*. There was fourteen times as much alkaline phosphatase (per mgm. protein) in the preparations from



Suggested pathways for the biosynthesis of triglycerides in subcellular fractions of intestinal mucosa rat intestine as in those from the rabbit. The relatively low incorporation obtained in the rat with α-glycerophosphate might therefore be due to the more rapid splitting of this compound to inorganic phosphate and glycerol. The glycerol cannot then be used for glyceride synthesis[6]. α-Monostearin, when supplemented with adenosine triphosphate, coenzyme *A*, glutathione and magnesium chloride, stimulated the incorporation of labelled palmitate to about the same extent as in the rabbit.

A more detailed report of this work will be published elsewhere.

We wish to thank Prof. A. C. Frazer for his interest and encouragement. We also thank the Medical Research Council and the University of Birmingham for grants supporting this work.

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[1] Frazer, A. C, Schneider, R., and Sammons, H. G., *Gastroenterologia*, 85, 146 (1956).

[2] Schneider, W. C., and Hogeboom, G. H., *J. Biol. Chem*, 183, 123 (1950).

[3] Hübscher, G, Dils, R. R., and Pover, W. F. R, *Biochim. Biophys. Acta* (in the press).

[4] Stein, Y., Tietz, A., and Shapiro, B., *Biochim Biophys. Acta*, 26. 286 (1957).

[5] Dawson, A. M., and Isselbacher, K J, *Fed. Proc*, 18, 212 (1959).

[6] Buell, G. C, and Reiser, R, *J Biol. Chem*, 234, 217 (1959).

[7] Kornberg, A, and Pricer, W E., *J. Biol. Chem*, 204, 329 (1953)

Presence of Sialic Acid in Connective Tissues

SIALIC ACID was first demonstrated in the submaxillary gland by Blix[1] and afterwards in many secretions of higher animals—in liver, ovomucin, meconium, colostrum, erythrocytes, nervous tissue and micro-organisms. Comprehensive reviews on sialic acids have been published by Zilliken and Whitehouse[2] and by Blix[3].

The presence of sialic acid in connective tissue has not so far been reported.

In the course of our research on chemical composition of epiphyseal cartilage[4] and particularly on the mucopolysaccharidic components[5], the presence of this acid in epiphyseal cartilage of young rabbits was demonstrated[6]. We have extended this study to other tissues in order to see whether a polysaccharide containing sialic acid was present in the ground-substance of all connective tissues.

The tissues were hydrolysed with 0·1 *N* sulphuric acid at 80° C. for 1 hr.; the hydrolysate was filtered on a sintered glass filter and chromatographed on 'Dowex 2 × 8' by Svennerholm's method[7]; after elution with 1 *M* acetate buffer *p*H 4·6 the eluate was tested by the resorcinol reaction[7] and sialic acid was identified by the absorption spectrum of the chromatic reaction.

To identify and determine quantitatively the sialic acid, we used as standard compound N-acetylneuramic acid isolated from sheep submaxillary gland and kindly supplied to us by Prof. G. Blix.

The presence of sialic acid was demonstrated in the following tissues: epiphyseal cartilage of young pigs (1·6 mgm. per gm. fresh tissue), rib cartilage of young rabbits (0·6 mgm./gm.), bovine cornea (0·4 mgm./gm.), bovine aorta (0·6 mgm./gm.) and bovine dental pulp (0·6 mgm./gm.); smaller quantities are also present in bone and dentin.

From these results we may conclude that sialic acid is a diffuse constituent of the polysaccharidic fraction of ground substance in connective tissues.

We thank Prof. G. Blix for his generous supply of N-acetylneuraminic acid.

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[1] Blix, G., *Hoppe-Seylers Z. physiol. Chem.*, 240, 43 (1936).

[2] Zilliken, F, and Whitehouse, M. W., "Adv. Carbohydrate Chem.", 13, 237 (1958).

[3] Blix, G., Fourth Int. Congr Biochem., Vienna, 1, 94 (1958)

[4] Castellani, A. A., and Castellani-Bisi, C, *J Biol. Chem.*, 233, 920 (1958)

[5] Castellani, A. A., Ferri, G., and Zambotti, V, *Boll. Soc. Ital. Biol. Sper* (in the press).

[6] Ferri, G., Castellani, A. A, and Zambotti, V, *Rend Ist Lomb Sci. Lett.*, 93, 214 (1959).

[7] Svennerholm, L., *Acta Chem. Scand*, 12, 547 (1958)

Configuration of Inositol Phosphate in Liver Phosphatidyl Inositol

THE hydrolysis of synthetic glycerol 1-(inositol 2-phosphate)[1,2] has recently been compared with that of the glycerylphosphoryl inositol isolated from hydrolysates of liver phosphatidyl inositol[3]. The results suggest that the phosphate group in the original lipid is attached to the 1- or 2-hydroxyl of the *myo*inositol. Inositol 1-phosphate should be optically active, while inositol 2-phosphate should not.

Previous work[4,5] had suggested that the inositol monophosphate produced on hydrolysis of ground-nut or soya phosphoinositides was optically inactive. It was therefore concluded[6] that phosphatidyl inositol has the 2-phosphate structure. However, Pizer and Ballou[7] have now provided evidence that the inositol monophosphate from soya phosphoinositide has a small but definite optical activity ($[\alpha]^{25}_{589}$, + 3·4° for the *cyclo*hexylamine salt,' −9·8° for the free acid). This called for a re-investigation of the earlier results.

One of the problems in determining the rotation of inositol monophosphate from soya lipids is that there are contaminating sugar phosphates which are strongly dextro-rotatory. A small amount of such an impurity could easily mask the low inositol monophosphate rotation. The soya inositol monophosphate, prepared as before[5], has therefore been purified by passage through three successive 'Dowex 1' columns, two using the formic acid–ammonium formate system and one using the sodium borate–ammonium formate system[3]. It was finally crystallized as a *cyclo*hexylamine salt and shown to be optically active. Since the rotation was quite small, determinations were made at several different wave-lengths. The activity increased considerably at lower wave-lengths. Molecular rotation was as follows: +12° (600 mμ), +18° (500 mμ), −32° (400 mμ), −54° (350 mμ), −89° (300 mμ). Calculations are based on the di*cyclo*hexylamine salt. Using a similar salt of inositol monophosphate produced by enzymic degradation of phytic acid and believed to be the 2-phosphate[5], values of zero (within the error of the instrument) were obtained at all the above wave-lengths. Soya inositol monophosphate, as the *cyclo*hexylamine salt, had $[\alpha]^{25}_{589}$ + 3·1°. All measurements were made in water.

The soya phosphoinositide therefore seems to have an inositol 1-phosphate structure. We wished to confirm this for liver phosphatidyl inositol, which may not have the same structure, and so an entirely different method was used. Acid or alkaline hydrolysis would be expected to produce the same mixture of inositol 1- and 2-phosphates whether the original lipid had the 1- or 2-structure. By analogy with the action of diesterases on polynucleotides, it seemed probable that enzymic hydrolysis of phosphatidyl inositol would give only one isomer and avoid migration problems. An enzyme releasing inositol monophosphate from this lipid had already been isolated in our laboratory[8]. Samples of the barium salt of inositol monophosphate obtained from the enzymic digest were chromatographed on paper in 6 : 3 : 1 *n*-propanol/ammonia/water, after removal of barium ions with 'Zeo-Karb 225'. Inositol 1- and 2-phosphates are separable in this solvent[7,9] if the papers are not overloaded. Comparison with authentic 2-phosphate and a mixture of 1- and 2-phosphates produced by acid-catalysed migration[9] showed that the enzyme released inositol 1-phosphate. A trace of the 2-compound was seen, probably produced during the isolation.

This, together with the evidence from the hydrolysis of synthetic glycerol 1-(inositol 2-phosphate), points to the inositol 1-phosphate structure for liver phosphatidyl inositol. Hanahan and Brockerhoff[10] recently studied the periodate oxidation of glycerylphosphoryl inositol and reached the same conclusion.

A more detailed account of this work will be published elsewhere.

We are indebted to Dr. W. Klyne of the Postgraduate Medical School, London, for the rotatory dispersion studies. We wish to thank Prof. A. C. Frazer for his continued interest. Miss M. Bason provided valuable technical assistance.

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[1] Davies, J. H., and Malkin, T., *Nature*, 184, 789 (1959).
[2] Ellis, R. B., and Hawthorne, J. N., *Nature*, 184, 790 (1959).
[3] Hawthorne, J. N , and Hubscher, G , *Biochem. J* , 71, 195 (1959)
[4] Iselin, B., *J. Amer. Chem. Soc* , 71, 3822 (1949).
[5] Hawthorne, J. N., *Biochim Biophys. Acta*, 18, 389 (1955)
[6] Hawthorne, J. N., and Hubscher, G., *Biochem J.*, 72, 10,*P* (1959).
[7] Pizer, F. L , and Ballou, C. E , *J. Amer. Chem Soc.*, 81, 915 (1959).
[8] Kemp, P , Hubscher, G , and Hawthorne, J. N., *Biochim. Biophys Acta*, 31, 585 (1959)
[9] Posternak, T., *Helv Chim. Acta*, 42, 390 (1958)
[10] Hanahan, D J , and Brockerhoff, H , *J. Amer. Chem Soc* , 81, 2501 (1959).

Antigen–Antibody in Liquid Two-Phase Systems : a Method for studying Immunological Reactions

THE behaviour of proteins[1–3], viruses[4–7] and cell particles[8,9] in aqueous polymer two-phase systems has recently been described. These phase systems consist of water and two water-soluble polymers. Mixtures in water of dextran and methylcellulose, or dextran and polyethylene glycol, are examples of such phase systems. The important characteristic of these systems is their high water content, so that most biochemical substances can be handled in them without denaturation. Each protein or virus particle is distributed in such phase systems in a characteristic manner so that a reproducible partition coefficient is found (partition coefficient, K = concentration of the partitioned substance in the top phase divided by the concentration of the same substance in the bottom phase) The partition coefficient of a particle in a given phase system depends mainly on the surface property of the particle and its size. The larger the size of particle the more unilateral is the distribution. Changes in the size of a protein molecule are reflected in a change in the partition coefficient For example, the dissociation of *Helix pomatia* hæmocyanin at certain pH values may be readily followed by measuring its partition coefficient in a dextran–methylcellulose system. Other changes in the structure of a protein molecule, such as the denaturation of serum albumin at low pH values, may cause the protein to move from one phase to the other.

In an immunological reaction the antigen and the antibody combine to form a complex which has a size larger than either of the reactants and possibly also has somewhat different surface properties. It was therefore of interest to study such a reaction in a two-phase system. The present communication describes experiments in which the reaction between the protein phycoerythrin and its rabbit antiserum in the phase systems dextran–methylcellulose and dextran–polyethylene glycol has been studied. Phycoerythrin was chosen as an antigen because it is coloured and can easily be measured by its strong light absorption at 540 mμ. In addition, its behaviour in the phase system has been previously studied in detail[1,3].

A typical experiment in the dextran–polyethylene glycol system was carried out in the following way. 0 5 ml. of an antigen solution and 1 ml. of diluted antiserum, both dialysed against 0·1 M phosphate buffer pH = 6 8, were mixed and allowed to stand at room temperature for 1 hr. A mixture was then added which contained dextran ('Dextran 500' from Pharmacia, Uppsala, Sweden) and polyethylene glycol ('Carbowax 6000', from Carbon and Carbide Chemicals, U.S.A.), and phosphate buffer pH = 6·8, in such proportions that, after mixing with the antigen–antibody solution, the concentration in dextran was 7 per cent w/w, polyethylene glycol 4·4 per cent w/w and the phosphate buffer 0·1 M. Mixing was then accomplished by inverting the tubes several times. Blanks containing antigen + normal sera and antigen only were run in parallel. The tubes were allowed to stand 2 hr. for phase separation.

In the dextran–polyethylene glycol system with 0 1 M phosphate buffer pH = 6·8, phycoerythrin is distributed in favour of the top phase with a K-value of about 5 while γ-globulin is distributed in favour of the bottom phase with a K-value of about 0·3. Thus the major portion of the antigen and the antibody when separate go to different phases in this system.

It could then be observed that in those tubes which contained antigen and antibody together, a red-coloured precipitate had collected at the interface. This occurred even in those tubes where the antigen was present in excess, that is, when the same proportion of antigen–antiserum mixed without the phase system did not show a precipitate. Thus, the presence of soluble antigen–antibody complexes had been visualized by a precipitation at the interface. Normal serum did not affect the distribution of the antigen.

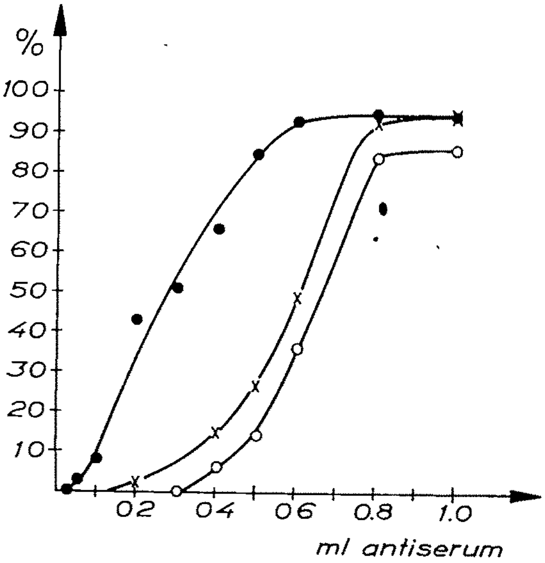

Fig 1 Immunological reaction between a constant amount of phycoerythrin (0·5 ml with E(1 cm., 540) = 2·16) and its rabbit antiserum in a two-phase system of dextran and polyethylene glycol at room temperature (filled circles), and by ordinary precipitin tests at room temperature (open circles) and at + 37° C. (crosses) For the phase system tests the reduction in the extinction of the top phase in per cent of the extinction of the top phase of the blank is plotted as the ordinate. For the ordinary precipitin tests the amount of antigen precipitated by centrifugation as a percentage of the total antigen is plotted as the ordinate

In order to estimate the amount of antigen precipitated at the interface, samples were drawn from the top phase, diluted with half their volumes, and the extinction at 540 mμ was measured. A reduction in the extinction indicates that antigen had precipitated, and this was expressed in per cent of the antigen in the blank containing no serum. Fig. 1 shows the result of such an experiment.

When a study was made of the effect on the immunological reaction of dextran or polyethylene glycol at the same concentrations as in their respective phases, it was found that they increased the precipitation in the tubes containing excess antigen. It is therefore probable that in the dextran–polyethylene glycol system the adsorption of precipitate at the interface is almost entirely due to a decreased solubility of the antigen–antibody complex in the phases. However, the fact that one part of the complex, the antigen, has more affinity for the top phase and that the other part, the antibody, has more affinity for the bottom phase may possibly contribute also toward increasing the adsorption of the complex at the interface.

An experiment in the dextran–methylcellulose system was carried out in the following way. 0·2 ml. antigen and 0·8 ml antiserum at different dilutions were mixed and allowed to stand at room temperature for 2 hr. A polymer mixture containing dextran (dextran with a limiting viscosity number of 0·68 dl./gm.) and methylcellulose ('Methocel 4000', from Dow Chemical Co., U.S.A.) in such proportions that after the addition to the antigen–antibody mixture a concentration in dextran of 0·64 per cent (w/w) and a concentration in methylcellulose of 0·56 per cent (w/w) was obtained. The final system also contained 0·15 M sodium chloride and 0·01 M phosphate buffer $pH = 6{\cdot}8$. This phase system is similar to system B of ref. 1 but contains a smaller bottom phase.

In the dextran–methylcellulose system both phycoerythrin and γ-globulin are separately distributed rather evenly, although slightly in favour of the bottom phase (K value about 0·9)[1]. In this system it was found that protein molecules and virus particles are distributed mainly according to their surface areas[4]. The appearance of the tubes containing antigen and antibody together was as follows. In the tubes with the largest amounts of antiserum a precipitate had fallen to the bottom of the tube. These tubes contained antigen and antibody in such proportions that when they were mixed in the absence of the phase system a precipitate was also formed. When antigen was in excess no precipitate was observed, but almost all the colour was in the bottom phase. In the presence of normal serum the colour was evenly distributed. Thus, the formation of soluble antigen–antibody complex had been visualized, in this system, by a unilateral distribution of the antigen colour.

The extinction of the top phase was measured as described above, and in Fig. 2 the reduction of the extinctions is expressed in per cent of the extinction of the blank containing antigen and normal serum.

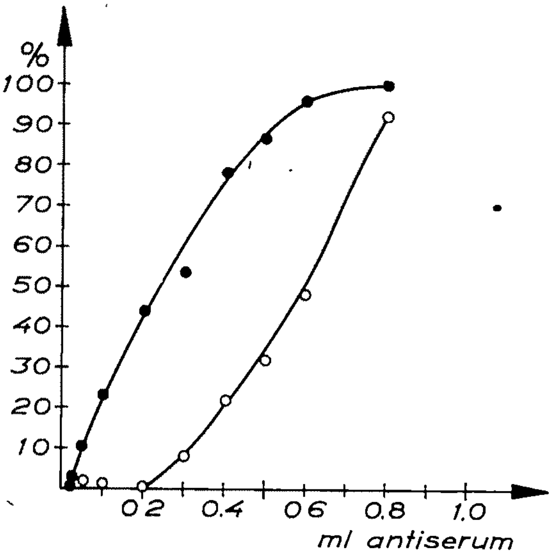

Fig. 2. Immunological reaction between a constant amount of phycoerythrin (0·2 ml. with E(1 cm., 540) = 5·0) and its rabbit antiserum in a two-phase system of dextran and methylcellulose at room temperature (filled circles) and by ordinary precipitin tests at room temperature (open circles). The ordinates are plotted as in Fig. 1

The two polymers, dextran or methylcellulose, did not themselves have any effect on the immunological reaction when they were present in the same concentrations as in their respective phases.

It is therefore probable that the unilateral distribution of the antigen is in a large part due to the size of its complexes with the antibody. According to previous findings, large protein molecules such as hæmocyanin and virus particles have a rather uneven distribution in the dextran–methylcellulose system used here[1,4].

Figs. 1 and 2 show that an increased sensitivity in measuring the antigen–antibody reaction in the antigen excess region has been achieved by both these systems. It is therefore suggested that aqueous polymer phase systems may be used with advantage in the study of immunological reactions, particularly those which do not give rise to a precipitate in ordinary tests.

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[1] Albertsson, P. Å., *Nature*, **182**, 709 (1958).
[2] Albertsson, P. Å., and Nyns, Ed. J., *Nature*, **184**, 1465 (1959).
[3] Albertsson, P Å , and Nyns, Ed. J. (in preparation).
[4] Albertsson, P. Å., and Frick, G., *Biochim. Biophys. Acta* (in the press)
[5] Frick, G., and Albertsson, P. Å., *Nature*, **183**, 1070 (1959).
[6] Wesslén, T , Albertsson, P. Å., and Philipson, L., *Archiv Virusforsch.* (in the press).
[7] Philipson, L , Albertsson, P. Å., and Frick, G. (in preparation).
[8] Albertsson, P. Å., *Biochim. Biophys. Acta*, **27**, 378 (1958).
[9] Albertsson, P. Å , Hanzon, V., and Toschi, G , *J. Ultrastructure Res* , **2**, 366 (1959).

Presence of Metanephrine and Normetanephrine in Normal Human Urine

IN 1957, Axelrod reported that epinephrine and norepinephrine were O-methylated at the C-3 position, giving metanephrine (3-O-methylepinephrine) and normetanephrine (3-O-methylnorepinephrine), respectively. in the rat[1]; in addition, he found that these compounds were normally excreted in rat urine[2]. The excretion of normetanephrine in the urine of humans with pheochromocytoma has also been reported[3]. More recently, the importance of the pathway for 3-O-methylation of epinephrine has been confirmed in man[4]. This report concerns the identification of metanephrine and normetanephrine of endogenous origin in the urine of normal human males.

Preliminary attempts to isolate these compounds were not successful because free metanephrine and normetanephrine are present in small amounts and are labile. Since the major portion of these metabolites was found to be excreted in a conjugated form, the conjugates were isolated by chromatography on ion-exchange resin[5], freed by hydrolysis, and identified.

A tracer amount (less than 0·018 μM) of a conjugate of tritiated metanephrine sulphate (3-O-methyl-7-^{3}H-epinephrine-4-O-sulphate), prepared biosynthetically, was added to 100 ml. portions of five individual urine specimens from normal human males; this amount of ^{3}H-metanephrine sulphate was too small to give spots on the chromatograms. These specimens were filtered and chromatographed on 1 × 10 cm columns of 'Dowex 1', acetate form. Distilled water (200 ml.) and 3-100 ml. portions of 0·2 N acetic acid were passed through each of the columns; the effluent was collected in 100 ml. fractions.

The radioactivity was eluted in the 0·2 N acetic acid fractions; these fractions from each individual column were combined and lyophilized. The resulting residues were redissolved in water. No substances in these urine extracts gave the phenolic reactions of metanephrine or normetanephrine prior to hydrolysis. Aliquots from the concentrated solutions of urine extracts were hydrolysed by heating with 1 N hydrochloric acid in a water bath at 37° C. for 48 hr. Approximately 2 per cent of each of the hydrolysed extracts was chromatographed on Whatman No. 1 filter paper in four different solvent systems: (*a*) *n*-butanol/glacial acetic acid/water (80 : 20 : 20); (*b*) *iso*-propanol/7 N ammonia (80 : 20); (*c*) benzene/propionic acid/water (40 : 40 : 20), and (*d*) *n*-butanol/*iso*-propanol/15 N ammonia (80 : 30 : 20). The urinary metabolites gave colour reactions with diazotized sulphanilic acid, diazotized *para*-nitroaniline and N-2,6-trichloro-*p*-quinoneimine that were identical with authentic metanephrine and normetanephrine.

The colour reactions, migration on paper chromatograms, and behaviour on ion exchange resins indicate that endogenous metanephrine and normetanephrine are excreted by normal human males in the absence of pheochromocytoma or of administration of exogenous epinephrine. The metabolites are excreted mainly as conjugates; detailed evidence to be presented elsewhere indicates that these are sulphoconjugates in humans. Although much valuable information has been obtained by analysis of urinary epinephrine and norepinephrine, these compounds represent only a small percentage of the total endogenous production. Since urinary metanephrine and normetanephrine, free and conjugated, and 3-methoxy-4-hydroxymandelic acid represent the major portion of exogenous, and presumably endogenous, epinephrine and norepinephrine[1,2,4], analysis of these O-methylated metabolites appears to offer promise of a more valid estimate of endogenous production of these important hormones.

We wish to thank Dr. Julius Axelrod and Dr. Seymour S. Kety for many helpful discussions during the course of this study.

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[1] Axelrod, J., *Science*, **126**, 400 (1957).
[2] Axelrod, J., Senoh, S., and Witkop, S., *J Biol. Chem* , **233**, 697 (1958).
[3] LaBrosse, E. H , Axelrod, J , and Sjoerdsma, A , *Fed. Proc.*, 17, 386 (1958)
[4] LaBrosse, E H , Axelrod, J., and Kety, S., *Science*, **128**, 593 (1958).
[5] Kirchner, N., Goodall, Mc C , and Rosen, L., *Proc. Soc. Exp. Biol. Med.*, **98**, 627 (1958).

ANIMAL PHYSIOLOGY

Reserpine and the Levels of Serotonin and Norepinephrine in the Brain

As an outgrowth of the results obtained in a number of laboratories, there appeared an explanation for the effect of reserpine which was based on the existence of reduced levels of serotonin and norepinephrine in the brain at a time when no reserpine was detectable[1]. Most of these methods suffered from the experimental use of massive doses of reserpine, which rendered the animal quite immobile. Subsequent work, using much lower doses of reserpine labelled with tritium and carbon-14, demonstrated that reserpine was present in the brains of guinea pigs at an unchanging concentration over the entire period of observable pharmacological effects[2]. From these investigations it was not possible to demonstrate any correlation between the intensity of effect of the drug and its concentration in the brain. With these

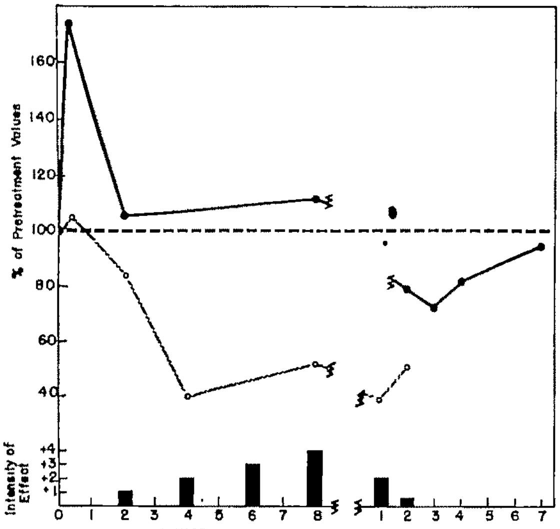

Fig. 1. Changes in levels of brain serotonin and catecholamine in the guinea pig at various times after a subcutaneous injection of 0·1 mgm./kgm. of reserpine. —●—●— catecholamines; —○— —.—○—, serotonin

facts in mind it was decided to determine if the changes in the levels of brain serotonin and/or norepinephrine could be correlated with the observable pharmacological effects.

As in the experiments with the labelled reserpine, the drug was administered subcutaneously at a dose of 0·1 mgm./kgm. to female guinea pigs weighing approximately 300 gm. The animals were killed by decapitation at the times shown in Fig. 1. The values in the graphs represent the average from 8 animals, except in the case for 2- and 3-day norepinephrine analyses, which are derived from 12 animals. Catecholamine analyses were carried out on ether-washed trichloracetic acid extracts according to Weil-Malherbe and Bone's method[3], and using a Klett fluorimeter. Careful adjustment of *p*H was necessary before addition of the tissue extracts to alumina columns. Serotonin was measured by Bogdanski's method[4], using an Aminco spectrophotofluorimeter.

20 min. after the injection of reserpine, the animals showed no gross drug effects and brain serotonin-levels did not vary statistically from control values. Catecholamine-levels, however, showed a 74 per cent increase over controls. After 2 hr. the animals began to show signs of drug action in the form of a very mild ptosis. Serotonin-levels decreased by about 16 per cent and catecholamine-levels had returned essentially to control values. The pharmacological effects increased in intensity, so that after 4 hr. the ptosis had become more severe and miosis was evident. Serotonin-levels had by this time reached 60 per cent of control values.

8 hr. after injection, the gross pharmacological effects had reached maximum intensity, while serotonin- and catecholamine-levels did not change from the previously measured value. The effects of the drug began to decrease in intensity, so that after two days almost all the symptoms had disappeared. At this time the serotonin-levels had not changed significantly from the 4-hr. value, while the catecholamine-levels had begun to fall, reaching 79 per cent of control values. After three days, the catecholamine value had fallen to 72 per cent of controls, and by seven days was essentially back to control-levels. This fall after two and three days, as well as the rise after 20 min., were found to be highly significant at the 1 per cent level, as determined by analysis of variance and the D. B. Duncan multiple range test[5] (we wish to thank Mrs. H. D. Slywestrowicz for the statistical analyses of the results).

Heart catecholamines increased by 29 per cent in 20 min. and 45 per cent in 2 hr. and then fell to control values at 8 hr. These levels continued to fall, reaching 50 per cent of controls by 24 hr. and remaining there for seven days. Spleen catecholamines also showed the increase at 20 min., reaching almost twice the control values.

These results constitute the first demonstration of a rise in catecholamine content following the administration of reserpine. By the use of selective filters and different times and heats of reaction during the condensation of catecholamine with ethylenediamine, it was established that epinephrine contributed little if any to the fluorescence which developed. While some of this fluorescence is undoubtedly due to dopamine, it had been shown that the concentration of this catecholamine was also reduced by large doses of reserpine[6].

In any event, these results have shown that the changes in the levels of catecholamines and serotonin following reserpine do not necessarily occur in the same direction or to the same degree. In addition, they demonstrate that under the conditions of the experiment, where a pharmacological rather than a toxicological dose was administered subcutaneously, the intensity of the gross observable effects of reserpine could not be correlated with the levels of amines in the brain. It is also apparent that neither the changes in amine-levels nor the intensity of drug effect could be correlated with the concentration of reserpine in these brains[2]. Until it becomes possible to understand the manner in which these amine-levels are maintained, it will be difficult to describe the manner in which they are altered by drugs. It may be necessary to look at mechanisms other than release of amines to explain adequately the scope of action of this potent alkaloid.

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[1] Brodie, B. B., and Shore. P. A., *Ann. N.Y. Acad. Sci.*, 66, 631 (1957).

[2] Sheppard, H., Tsien, W. H., Plummer, A. J., Peets, E. A., Giletti, B. J., and Schulert, A. R., *Proc. Soc. Exp. Biol Med.*, 97, 717 (1958).

[3] Weil-Malherbe, H., and Bone, A. D., *Biochem. J.*, 51, 311 (1952).

[4] Bogdanski, D. F., Pletscher, A., Brodie, B. B., and Udenfriend, S., *J. Pharmacol. Exp. Therap.*, 117, 82 (1956).

[5] Duncan, D. B., *Biometrics*, 2, 1 (1955).

[6] Carlsson, A., Lindqvist, M., Magnusson, T., and Waldeck, B., *Science*, 127, 471 (1958).

Effect of Inorganic Sulphate on Molybdenum Excretion by a Marsupial

Dick and Bull[1] published the first record of a copper–molybdenum interaction in sheep and cattle in which copper stored in the liver is reduced by increasing molybdenum intake. Later, Dick[2] showed that inorganic sulphate has to be ingested with molybdenum before this element can exert a limiting effect on the storage of copper in the liver of sheep Afterwards, Wynne and McClymont[3] found that it was possible to produce experimental hypocuprosis by feeding penned sheep intake levels of copper, molybdenum and inorganic sulphate of the same order as those which occur naturally in pastures of New South Wales, where sheep develop hypocuprosis. Spais[4] has described a similar 'induced' copper deficiency in flocks of sheep grazing on coastal areas of Thessalonika, Greece, where food plants have normal copper but high molybdenum and inorganic sulphate levels.

The quokka, *Setonix brachyurus*, Quoy and Gaimard, is found in a natural population on Rottnest[5], a small island 12 miles west of Fremantle, composed of wind-blown sand (æolionite). Following the work of Moir *et al.*[6], who showed that the quokka has ruminant-like digestion, it was decided to embark on a project to determine whether trace-element deficiencies played any part in the biology of the animal on the island, because, due to their origin, the soils were likely to be deficient, especially in copper and cobalt. Furthermore, it had been found that some animals showed a marked seasonal anæmia which has since been documented by Shield[7].

Analysis of the plants eaten by the quokka on Rottnest has shown that copper-levels are extremely low (usually less than 3 p.p.m. on a dry-weight basis), molybdenum-levels are moderate (up to 8 p.p.m. dry-weight basis) and inorganic sulphate-levels are high (up to 2·3 per cent on a dry-weight basis). The first experimental step was to determine the effect of inorganic sulphate on the excretion of molybdenum. An experiment similar to that used by Dick[8] on sheep was carried out. Two pairs of adult male quokkas, placed separately in metabolism cages, were given food and water *ad lib.* and a 24-hr. collection of urine was made. At the end of this period, blood samples were taken. The animals were then given a daily drench of ammonium molybdate; the total daily intake of molybdenum of each animal from the food and drench was 600 μgm. At the end of a week a 24-hr. collection of urine and a blood sample were taken from each animal. One animal of each pair was then given a drench of ammonium sulphate containing 1·25 gm. sulphate and the other a drench of ammonium chloride containing the same amount of chloride. Daily drenching with molybdate was continued and blood and urine collection was taken for the two following 24-hr. periods. All samples were analysed for copper and molybdenum.

The results of an analysis of variance were only significant ($P < 0.01$) for the changes in blood and urine molybdenum which occurred after drenching with inorganic sulphate. Table 1 shows that in both groups the mean molybdenum-levels of whole blood rose after drenching with molybdate solution for one week. Drenching with sulphate caused a decrease in whole-blood molybdenum-levels to pre-treatment levels in 24 hr. and there was a corresponding increase in the total excretion of molybdenum in the urine for this period. Drenching with chloride had no effect on the excretion of molybdenum in the urine. Blood copper-levels and total copper excreted in the urine of both groups of animals remained unchanged. There was no significant increase in urine volume in the sulphate group after drenching with ammonium sulphate or in the chloride group after drenching with ammonium chloride.

Table 1. Changes in Blood Molybdenum-levels and Total Molybdenum excreted in the Urine induced by a Single Dose of Inorganic Sulphate

(The dotted line indicates the time of drenching with sulphate and chloride solutions)

		Urine volume (ml.)	Blood molybdenum (μgm./ml.)	Blood copper (μgm./ml.)	Urine molybdenum (total μgm.)	Urine copper (total μgm.)
Sulphate group	Day 0	248	0·05	0·66	1·9	7·8
	Day 7	201	0·20	0·70	56·0	6·5
	Day 8	182	0·08	0·64	141·0	8·4
	Day 9	139	0·11	0·65	56·0	6·8
Chloride group	Day 0	201	0·04	0·57	4·1	7·3
	Day 7	135	0·17	0·66	90·0	4·3
	Day 8	215	0·20	0·69	77·5	6·1
	Day 9	141	0·16	0·68	89·2	7·1

The results indicate that inorganic sulphate has an effect on molybdenum excretion in the quokka similar to that in the sheep.

An experiment to test the long-term effect of increased intake-levels of molybdenum and inorganic sulphate on blood copper-levels and the storage of copper in the liver of the quokka is at present being carried out. Preliminary results show that an intake-level of molybdenum and inorganic sulphate comparable to field-levels at Rottnest, exert a depressing effect on blood copper-levels. This work will be published on completion.

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[1] Dick, A. T., and Bull, L. B., *Aust. J. Vet.*, 21, 70 (1945)

[2] Dick, A. T., *Nature*, 172, 637 (1953)

[3] Wynne, K. N., and McClymont, G. L., *Nature*, 175, 471 (1955).

[4] Spais, A., Pathological Clinic of the Veterinary School, University of Thessalonika (1956)

[5] *J. Roy. Soc. West Australia*, 42, Pt. 3, edit. by Hodgkin, E. P., and Sheard, K. (1959)

[6] Moir, R. J., Somers, M., Sharman, G. B., and Waring, H., *Nature*, 173, 269 (1954).

[7] Shield, J. W., Ph.D. thesis, University of Western Australia (1958).

[8] Dick, A. T., *Aust. Vet. J.*, 29, 18 (1953).

Minimum Dose of Diethylstilbœstrol required to stimulate the Phagocytic Activity of the Reticulo-Endothelial System

DIETHYLSTILBŒSTROL has been reported previously by Nicol *et al.* [1,2] to have a strong stimulative action on the phagocytic activity of the reticulo-endothelial system, and various steroids having high œstrogenicity have also been shown to have a similar action[3]. The present research was planned to determine the minimum dose of diethylstilbœstrol required to stimulate the reticulo-endothelial phagocytes and to ascertain its relationship to the œstrogenic potential of the substance.

The present experiments were carried out on 75 male white mice (*T.O.* Swiss strain) of 20–28 gm. body-weight. Groups of five animals were used for testing the various doses of diethylstilbœstrol. All the animals of each group received one subcutaneous dose of diethylstilbœstrol in 0·05 ml. of arachis oil daily for six days. The doses of diethylstilbœstrol ranged from 1 × 10^{-6} mgm. to 0·6 mgm. On the eighth day after the commencement of the treatment the phagocytic activity was measured by the rate of disappearance of a known amount of carbon from the circulating blood[4], the procedure used being that described in previous communications[2,3], and the total body phagocytic activity or phagocytic index being denoted by the symbol K.

Twenty of the animals were used as controls and were given 0·05 ml. arachis oil once daily for six days; the phagocytic activity was then measured on the eighth day by the carbon method referred to above. The arachis oil controls showed an average phagocytic index or K value of 15 ± 0·5.

The results of the different doses of diethylstilbœstrol used are shown in Table 1. Compared with the control values, it can be seen that definite stimulation of phagocytic activity does not occur until the daily dose of diethylstilbœstrol rises to 0·1 mgm. Thereafter, although stimulation is maintained, it does not increase in proportion to the dose. In contrast, the dose necessary to induce œstrus is 0·001 mgm. The minimum dose required to stimulate the reticulo-endothelial phagocytes is therefore about 100 times greater than the œstrogenic potential of the substance.

Table 1. EFFECT OF GRADUAL INCREASE OF DOSE OF DIETHYLSTILBŒSTROL ON THE PHAGOCYTIC ACTIVITY OF THE RETICULO-ENDOTHELIAL SYSTEM

Diethylstilbœstrol (daily dose × 10^{-6} mgm.)	Phagocytic index (K value)	Percentage change in body-weight
1	20 ± 2·1	+ 6·8
10	10 ± 1·3	+ 5·9
100	21 ± 2·9	+ 5·2
1 × 10^3	16 ± 1·8	+ 11·6
1 × 10^4	28 ± 1·7	+ 14·3
1 × 10^5	89 ± 13·2	+ 2·0
2 × 10^5	68 ± 21	− 18·5
3 × 10^5	67 ± 7·7	− 11·6
4 × 10^5	141 ± 38	− 8·5
5 × 10^5	96 ± 11·5	− 13·0
6 × 10^5	103 ± 19	− 10·5
Arachis oil controls in 20 animals	15 ± 0·5	—

It is also of interest to note the effect of increasing doses of diethylstilbœstrol on the body-weight of the animals. Generally, small doses increasing to 0·1 mgm. are accompanied by a gain in weight, but with doses above this level the animals constantly lose weight.

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[1] Nicol, T., Helmy, I. D., and Abou-Zikry, A., *Brit. J. Surg.*, **40**, 166 (1952).

[2] Nicol, T., Bilbey, D. L. J., and Ware, C. C., *Nature*, **181**, 1538 (1958).

[3] Bilbey, D. L. J., and Nicol, T., *Nature*, **182**, 674 (1958).

[4] Biozzi, G., Benacerraf, B., and Halpern, B. N., *Brit. J. Exp. Path.*, **34**, 441 (1953).

Fall in Hæmatocrit in Sheep after Treatment with Chlorpromazine and Dihydroergotamine

IT was recently shown[1] that the jugular hæmatocrit of sheep often fell considerably (by as much as 24 per cent of the initial value, mean = 12 per cent) when the animals were isolated from alarming environmental stimuli, or were subjected to gentle restraint ('holding') and soothing for 15–30 min., and that it fell regularly and to an even greater extent (up to 39 per cent of the initial value, mean = 30 per cent) during anæsthesia by pentobarbitonum. Evidence was obtained that these phenomena are a consequence of splenic relaxation, passive filling of the spleen, and segregation therein of an increased fraction of the total red cell mass in a state of dynamic equilibrium with the circulating blood. The remarkable fall in hæmatocrit after 'holding', which we view as emotional or psychological tranquillization leading to diminution of the sympatho-adrenal flux from the cerebral cortex, suggested that this might also occur as a result of pharmacological tranquillization by means of ataractic agents.

This has proved to be so. Intravenous injection of chlorpromazine (0·46–0·7 mgm./kgm.) was followed by rapid hæmatocrit fall, reaching within 60 min. minimum values which were 30 per cent less than the initial value and followed by gradual return to pre-injectional levels after some hours. Plasma specific gravity changes indicated that plasma dilution through water shift accounted for a small part of the hæmatocrit fall; when hæmatocrits were adjusted to eliminate the contribution of this factor it was found that the relative numbers of circulating erythrocytes in the jugular blood fell by 26 per cent. In splenectomized sheep erythrocytes in the jugular blood fell by only 3–4 per cent. Furthermore, in a sheep prepared on the previous day with ^{51}Cr-marked red cells, hæmatocrit fall and gradual return, following injection of chlorpromazine, were mirrored by reciprocal increase and gradual return of gamma-radiation surface counts over the splenic region. It therefore appears that the hæmatocrit fall in intact sheep after chlorpromazine is due mainly to increased retention of red cells in the spleen.

Chlorpromazine was shown to inhibit hæmatocrit rise after adrenaline or *nor*adrenaline intravenous injection. Inhibition was also demonstrated after previous intravenous injection of 'Dibenamine' (20 mgm./kgm.); but this adrenergic-blocking agent caused only a minor (3 per cent) decrease of hæmatocrit. On the other hand, after intravenous injection of dihydroergotamine (14–168 μgm./kgm.), with which inhibition of hæmatocrit rise after the two sympathomimetic catechol amines was also demonstrated, hæmatocrit fell quickly and profoundly (up to 24 per cent decrease), whereas in a splenectomized sheep injection of 1,680 μgm./kgm. was followed by a trivial fall (3 per cent decrease). However, this substance is known to have marked central effects[2,3] in addition to its adrenergic-blocking effects, whereas the adrenergic-blocking activity of 'Dibenamine' is more specific and it has little central effect[4]. Furthermore, intravenous injection of pentobarbitonum sodium, which prevents sympatho-adrenal discharge by blocking nervous impulses within the central nervous system[4], was followed by moderate but slow hæmatocrit fall (progressive 10–12 per cent decrease within 3 hr.) when given in a dose (10 mgm./kgm.) sufficient to cause mild sedation accompanied by muscular inco-ordination. It therefore appears that the splenic relaxation in sheep after chlorpromazine may be due mainly to central effects, and predominantly to its ataractic properties, causing emotional indifference to otherwise alarming environmental stimuli, rather than to direct adrenergic-blocking effects within the spleen.

Fall in hæmatocrit (18 per cent decrease) and its virtual abolition by splenectomy was also demonstrated in sheep after intramuscular injection of mepazine (2·3 mgm./kgm.). The hæmatocrit fall after chlorpromazine injection reported in dogs[5] is doubtless also due to increased retention of red cells in the dilated spleen. This effect of chlorpromazine and mepazine, and probably of many other tranquillizing drugs, may be expected in many animals in which the red-cell storage function of the spleen is important.

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[1] Turner, A. W., and Hodgetts, V. E., *Aust. J. Exp. Biol. Med. Sci.*, 37 (in the press).

[2] Nickerson, M., *Pharmacol. Rev.*, 1, 27 (1949).

[3] Rosenblueth, A., and Cannon, B., *Amer. J. Physiol.*, 105, 373 (1933).

[4] Nickerson, M., *Pharmacol. Rev.*, 11, 443 (1959).

[5] Hoe, C. M., and Wilkinson, J. S., *Vet. Rec.*, 69, 734 (1957).

PLANT PHYSIOLOGY

Auxin-induced Methylation in Maize

The transfer of methyl groups from methionine to the hot-water-soluble fraction of *Avena* coleoptile cell walls is increased by indoleacetic acid (IAA)[1]. Unlike most effects of indoleacetic acid, this increase occurs even when all expansion is osmotically prevented. Thus this process appears to be directly controlled by auxin and may constitute a portion of the mechanism whereby auxin induces elongation. If this were so, it would be expected that auxin would influence this transfer in all tissues which show an auxin-growth response. Maize coleoptile and mesocotyl tissues have now been examined for the presence of auxin-induced methylation.

Avena coleoptile sections for use as controls were obtained in the manner of Ordin *et al.*[1]. Sections of maize coleoptile and mesocotyl were obtained as follows. Maize seedlings were germinated in moist sand and allowed to grow in the dark for about 90 hr. Mesocotyls of 2·5–3·5 cm. in length were then selected and one 5-mm. section was excised from each mesocotyl using a double-bladed cutter. The apical incision was 1 mm. below the node Coleoptiles of 1·5–2·0 cm. in length were selected at the same time and a 5-mm. section was excised after removal of the apical 3 mm. The central leaves were removed from the coleoptile sections of *Avena* and maize. Each experimental treatment contained sufficient material to give about 10 mgm. dry cell wall. All manipulations were carried out under a weak red light.

Sections were incubated in 0·0025 *M* potassium maleate solutions (*p*H 4·8), ± 5 p.p.m. indoleacetic acid. The methyl donor used was methionine-methyl labelled with carbon-14 (Amersham, 0·145 mc./mgm.) in a concentration of 2–3 × 10^{-5} *M*. At the end of the incubation period, the amount of saponifiable methyl groups in the hot-water-soluble pectin was determined using the methods of Ordin *et al.*[1].

Table 1. The Effect of Auxin on the Transfer of Methyl Groups from Methionine-^{14}C to Wall Pectin of Various Tissues

Material	Methionine administered (c.p.m.)	Time (hr.)	Growth-rate IAA/control	5 p.p.m IAA	MeOH-^{14}C/10 mgm. cell wall (c.p.m.)
Avena coleoptile	3·5 × 10^6	5	2·5	±	1,145 620
Maize coleoptile	2·9 × 10^6	4	2·1	±	3,210 2,010
Maize mesocotyl	3·85 × 10^6	20	2·0	±	1,930 1,945

The results are shown in Table 1. An auxin treatment causes an increase in the transfer of methyl groups from methionine to wall pectin in maize coleoptiles as well as in *Avena* coleoptiles. No such increase occurs in maize mesocotyls even though growth is doubled by auxin The lack of auxin-effect may mean that transfer of methyl groups to hot-water-soluble pectin is not a necessary part of the mechanism of auxin-induced growth. Alternatively, since it is known that other substrates such as formaldehyde can furnish the methyl groups to pectin (Sato, C., unpublished work), this result may only be a reflexion of a marked preference of maize mesocotyls for some substrate other than methionine as the source of pectin methyl groups.

This work has been carried out in the laboratory of Prof. T. A. Bennet-Clark at the Botany Department, King's College, London, and has been supported by post-doctoral fellowship *CF*-7341-*C* from the National Cancer Institute, United States Public Health Service. I wish to express my appreciation to Prof. Bennet-Clark for his help and encouragement and to Mrs. M. W. Ware for advice on methods of obtaining maize coleoptile and mesocotyl sections.

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[1] Ordin, L., Cleland, R., and Bonner, J., *Proc. U.S. Nat. Acad. Sci.*, 41, 1023 (1955).

Certain Mitotic Effects of Kinetin, Gibberellic Acid, Indoleacetic Acid, and Maleic Hydrazide on the Root of *Allium cepa*

Treatment of intact roots of *Allium cepa* with kinetin (6-furfurylaminopurine) has been reported by Guttman[1] to cause an increase in the rate of division of the cells of the root tip. She found a similar stimulatory activity of kinetin in promoting division of *Paramecium caudatum*[2].

Guttman made her studies of *Allium cepa* roots on random fields of squash preparations. The following is a report of similar studies in which the counts of mitotic figures were made on serial longitudinal sections of the root tips. The results indicate a markedly inhibitory effect on mitosis when kinetin is applied to intact roots in the same manner and in similar concentrations.

Table 1 shows the mean number of mitotic figures present in all tissues of the distal 5 mm. of the root tip, in 10-μ sections from the centre of five roots which had been treated with 1 p.p.m. and with

Table 1. MEAN NUMBERS OF MITOTIC FIGURES PRESENT IN ALL TISSUES OF THE DISTAL 5 MM. OF *Allium cepa* ROOTS TREATED WITH GROWTH-REGULATING SUBSTANCES. VALUES REPRESENT THE MEANS OF 20 CENTRAL SECTIONS 10μ THICK

Days	Control	K	GA	IAA	MH	K+GA	K+IAA	K+MH	GA+IAA	GA+MH	IAA+MH
					1 p.p m.						
1	17·1	3·1	10·8	5·8	7·6	2·9	0·8	0·8	7·6	5·0	3·7
2	16·5	5·0	14·2	9·0	10·1	3·9	4·8	9·1	8·5	5·8	6·4
3	18·6	1·5	14·6	15·4	5·2	5·2	5·6	6·5	7·8	7·7	3·8
4	18·8	2·8	18·5	24·6	2·0	8·1	6·8	7·2	14·8	6·8	6·0
5	19·8	7·9	26·9	27·7	2·2	8·9	7·3	6·1	21·8	8·1	5·0
7	17·0	6·4	18·3	19·9	1·8	13·8	1·1	1·6	26·0	1·6	0
					10 p.p m.						
1	17·4	5·1	14·7	5·6	4·1	4·5	3·8	8·3	3·5	3·2	3·5
2	16·0	2·3	7·0	0·7	3·7	1·1	1·4	2·7	2·0	1·3	0
3	17·5	0·2	2·4	0·1	0	0	1·6	0·5	0·2	1·3	0

K, Kinetin; GA, gibberellic acid; IAA, indoleacetic acid; MH, maleic hydrazide

10 p.p.m. kinetin for the times indicated in Table 1. The value given is the average of twenty counts for each treatment.

Roots were also treated with 1 p.p.m. and with 10 p.p.m. gibberellic acid, and with similar concentrations of indoleacetic acid and maleic hydrazide, as well as with combinations of all of these in all possible pairs. Results are summarized in Table 1.

In their review of the literature on the gibberellins, Stowe and Yamaki[3] listed no studies of the effect of cell division on intact roots, but it has recently been shown to stimulate cell division in shoots (for example, Sachs and Lang[4]). In the present investigation, roots treated with gibberellic acid showed initial depression of mitosis followed by an after-effect of stimulation of mitosis. The pattern of mitotic activity was similar to but not so pronounced as that seen after treatment with indoleacetic acid. The effects of indoleacetic plus gibberellic acids were not additive, however. At 10 p.p.m., gibberellic was inhibitory to mitosis and after 3 days the roots appeared dead.

Roots treated with indoleacetic acid at a concentration of 1 p.p.m. show inhibition of their longitudinal growth for about 3 days, during which time the characteristic bulbous swelling or 'c-tumour' is produced just proximal to the root tip. After about 3 days, elongation of the root is resumed, and some workers have noted that the rate of elongation may then exceed that of control roots (for example, Thimann[5]). In the present work it was found that mitotic activity exactly paralleled this characteristic response in the rate of total elongation of the root. Initially there was inhibition of mitosis, followed by a return to the level of the control at about 3 days. At 5 days, during the time when there is stimulation of gross elongation, the number of mitotic figures seen also exceeded that of the control. At 10 p.p.m., indoleacetic acid markedly inhibited mitosis, and there was no later release from the inhibition. After 3 days the roots appeared dead.

Interactions of indoleacetic acid with each of the other substances used here have been reported. No consistent reversal or enhancement of the initial effect of indoleacetic acid was seen here, but only when this acid was combined with gibberellic acid was the tendency to an after-effect of stimulation seen.

Maleic hydrazide has been reported to reverse the effect of indoleacetic acid in various test reactions (for example, Leopold and Klein[6]). In the present work it was always inhibitory, and it did not reverse the action of any of the auxin-like substances used.

Some workers have reported the presence of mitotic aberrations in plant material treated with maleic hydrazide, whereas others have not found them. McLeish[7] found no chromosome breaks after treatment of onion root tips with maleic hydrazide, and Greulach and Atchison[8] found no mitotic aberrations in onion root tips so treated. In the root tips treated with 10 p.p.m. maleic hydrazide in the present investigation, about one-third of the mitotic figures seen showed aberrations, including fragmentation and changes which gave the chromosomes a beaded appearance.

Clowes[9] has suggested that there is a 'quiescent centre' in the area of the apical initials of some roots, in which the cells normally rarely or never divide. It was noted here that mitotic figures were very rare in the centre of the apical initial area of *Allium cepa*.

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[1] Guttman, R., *Chromosoma*, 8, 341 (1956).
[2] Guttman, R., and Back, A., *Nature*, 181, 852 (1958).
[3] Stowe, B. B., and Yamaki, T., *Ann. Rev. Pl. Physiol.*, 8, 181 (1957).
[4] Sachs, R. M., and Lang, A., *Science*, 125, 1144 (1957).
[5] Thimann, K. V., *Amer. J. Bot.*, 24, 407 (1937).
[6] Leopold, A. C., and Klein, W. H., *Physiol. Plantarum*, 5, 91 (1952).
[7] McLeish, J., *Heredity* (Supp.), 6, 125 (1953).
[8] Greulach, V. V., and Atchison, E., *Bull. Torrey Club*, 77, 262 (1950).
[9] Clowes, F. A. L., *J. Exp. Bot.*, 7, 307 (1956).

RADIOBIOLOGY

Isolation of Radioactive Vitamin B_{12} from Incubation Mixtures containing Glycine-2-^{14}C and Turkey Liver Homogenates

STUDIES carried out with tracer techniques indicate that α-carbon of glycine gives rise to a single-carbon moiety at the oxidation-level of formaldehyde and this combines with α-carbon of another molecule of glycine to give rise to serine[1]. However, no free formaldehyde *per se* but formic acid has been isolated from the reaction mixtures containing liver homogenates, glycine-2-^{14}C or serine-3-^{14}C[2,3]. Both *in vitro* and *in vivo* experiments, compounds of the type of hydroxymethyltetrahydrofolic acid are produced from glycine and serine[4,5] and this is strong indirect evidence for the formation of formaldehyde from α-carbon of glycine or β-carbon of serine.

Vitamin B_{12} appears to be involved in the biosynthesis of serine from glycine[6] in turkey liver homogenates and also reacts with formaldehyde to form complexes *in vitro*[7].

Table 1. INCORPORATION OF RADIOACTIVITY FROM GLYCINE AND SERINE INTO FORMALDEHYDE AND VITAMIN B_{12}

Amino-acid	Specific activity (c p.m./m.mole) ($\times 10^6$)	Vitamin B_{12} added (mgm.)	Incubation time (hr.)	Total formaldemethone isolated (mgm.)	Specific activity of $H^{14}CHO$ (c.p.m./m.mole)	Total activity of isolated vitamin B_{12} (c.p m./mgm.)
Glycine-1-^{14}C	3 6	20	4	33	0	0
Glycine-2-^{14}C	3 5	20	18	25	3,250	10
	3 5	0	18	26	2,520	—
Glycine-2-^{14}C	2·3	20	12	16	1,950	8
	2 3	0	12	32	1,200	—
Serine-3-^{14}C	5 2	20	4	32	2,200	1

The production of free formaldehyde from glycine in reaction mixtures containing turkey liver homogenates has been investigated and an attempt has been made to confirm the formation of vitamin B_{12}-formaldehyde complexes from these reaction mixtures.

Turkey poults fed diets deficient in vitamin B_{12} were killed at the age of 3 weeks and their livers were homogenized in cold conditions and frozen. An approximately known volume was used in each experiment. The buffer, *p*H 7·6, contained 11·98 gm. crystalline disodium hydrogen phosphate and 1·02 gm. potassium dihydrogen phosphate per litre. Serine (DL-serine-3-^{14}C) with an activity of 929 μc./m.mole was obtained from Nuclear, Chicago, Ill. The dimedon solution contained 0·4 gm. dimedon (5,5-dimethyl-1,3-*cyclo*hexanedione) per 100 ml. water. Glycine-2-^{14}C with an activity of 1 mc./m.mole was obtained from Research Specialities Co., Berkeley, Calif. Glycine-1-^{14}C had an activity of 1·59 mc./m.mole and was supplied by New England Nuclear Corp., Boston, Mass. 1 ml. of 37 per cent formaldehyde solution was diluted to 50 ml. with water; semicarbazide solution, 0·4 per cent in water; glycine and serine solutions, 20 mgm./ml.; trichloracetic acid, acetic acid and sodium hydroxide solutions, 10 per cent.

The method used for the isolation of formaldehyde was essentially the same as described by Frisell and Mackenzie[8]. Each of the two flasks used in these experiments contained the following solutions: 2 ml. buffer, 0 5 ml. inactive glycine, 3 ml. radioactive glycine, 1 ml. formaldehyde and 1 ml. semicarbazide. One of the flasks also contained 20 mgm. vitamin B_{12}. Then 2 ml. of liver homogenate was added and flasks were covered with 'Parafilm'. After incubation at room temperature for 4–18 hr., 6 ml. of trichloracetic acid solution was added to each flask and the solutions were filtered through Whatman filter paper No. 42. The protein on the filter paper was further washed with 5 per cent trichloracetic acid solution until practically free from the colour of vitamin B_{12}. The solution was neutralized with sodium hydroxide solution, using an internal or external indicator as necessary, and again made just acidic with acetic acid solution. The solutions were filtered again; 25 ml. of dimedon solution was added to each flask and kept overnight in the refrigerator when formaldemethone separated out (mol. wt. 292). This was filtered, washed with water and recrystallized from dilute alcohol after treatment with carbon to a constant activity.

In this way, experiments were tried with glycine-1-^{14}C, glycine-2-^{14}C and serine-3-^{14}C.

Vitamin B_{12} was isolated from the solutions after filtration of formaldemethone essentially by the method of Fantes *et al.*[9]. 'Norit' carbon was used to adsorb vitamin B_{12} and the carbon was washed with 200–300 ml. of water and then with 50 ml. of 2 per cent phenol solution and again with water. Vitamin B_{12} was eluted from carbon with hot 65 per cent ethanol. The alcoholic solution was concentrated under reduced pressure, and residue was diluted with 50 ml. water and again adsorbed on 'Norit' carbon. The washing of 'Norit' and elution of vitamin B_{12} was repeated. The alcoholic solution was again evaporated to dryness under reduced pressure and the residue was dissolved in the minimum amount of water and acetone was added until a turbidity appeared. The solution was left in freezer overnight and crystals of vitamin B_{12} were obtained. Crystallization of vitamin B_{12} was repeated three more times.

Vitamin B_{12} was dissolved in dilute alcohol and streaked as a band on Whatman filter paper No. 2*MM*; glycine and serine were also spotted on the paper and it was chromatographed for 48–72 hr. in ascending butanol/acetic acid/water, 4 : 1 : 1. After drying the paper, the section of it containing glycine or serine was sprayed with ninhydrin and developed. The amino-acids moved far ahead of vitamin B_{12} and sections of paper corresponding to vitamin B_{12} were cut and eluted with water. The eluate was evaporated in polyethylene planchets and the radioactivity was determined.

Samples of formaldemethone were converted to carbon dioxide by wet combustion, precipitated as barium carbonate, filtered on filter paper disks and counted under a windowless gas flow counter. Proper corrections were made for self-absorption.

In some cases, the samples were plated in infinitely thin layers on polyethylene planchets and counted. No correction was made for self-absorption.

Preliminary experiments were carried out in which vitamin B_{12} was allowed to react with glycine-1-^{14}C, glycine-2-^{14}C and formic acid in absence of any liver homogenates. The procedure has been reported earlier[7]. In no case was any radioactivity found to be associated with vitamin B_{12}. This observation is in sharp contrast to the reaction of formic acid with tetrahydrofolic acid, where formyltetrahydrofolic acid is obtained.

Vitamin B_{12} isolated from reaction mixtures containing glycine-2-^{14}C, or serine-3-^{14}C, vitamin B_{12} and liver homogenates was found to be radioactive (Table 1). No radioactivity was associated with vitamin B_{12} incubated with glycine-1-^{14}C and liver homogenates.

Glycine-1-^{14}C is metabolized to yield labelled carbon dioxide and this is not exchanged with any carbon of vitamin B_{12}. For this reason, the vitamin B_{12} isolated from the reaction mixture was non-radioactive.

On the other hand, glycine-2-^{14}C and serine-3-^{14}C both yield labelled formaldehyde, as is clear from Table 1, where the specific radioactivities of formaldehyde isolated are given. Previous workers had failed to obtain any free formaldehyde because it might be easily oxidized to formic acid. The addition of some inactive formaldehyde helped to trap radioformaldehyde produced. If no liver homogenates were used, about 69 mgm. of formaldemethone was isolated. But with liver homogenates about 32 mgm. of this compound was isolated; the remaining formaldehyde must have been oxidized.

Also, formaldehyde isolated from the reaction mixtures containing vitamin B_{12} appeared to be slightly more radioactive than without it. This radioformaldehyde may have combined with vitamin B_{12} to give vitamin B_{12}–formaldehyde complexes. That radioactivity in vitamin B_{12} was not due to any chance contamination with amino-acids was proved by paper chromatography.

No definite information can be given about the molar ratios of vitamin B_{12} and formaldehyde in these complexes. In studies with glycine-2-^{14}C, each mole of vitamin B_{12} appears to be in combination with 4 or more moles of labelled formaldehyde. This calculation is based on a molecular weight of vitamin B_{12} of 1,356. But the ratio for the complex isolated from serine-3-^{14}C studies is very different. Here 2 moles of vitamin B_{12} appear to be in combination with 1 mole of labelled formaldehyde.

The isolation of vitamin B_{12}-formaldehyde complexes both *in vitro* and in present experiments suggests that these should be further investigated with respect to any role in serine–glycine interconversions.

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[1] Sakami, W., "Amino Acid Metabolism", ed. McElroy, W. D., and Glass, B., 658 (Johns Hopkins Press, Baltimore, 1955).

[2] Siekevitz, P., and Greenberg, D. M., *J. Biol. Chem.*, **180**, 845 (1949); **186**, 275 (1950).

[3] Mackenzie, C. G., "Amino Acid Metabolism", ed. McElroy, W. D., and Glass, B., 684 (Johns Hopkins Press, Baltimore, 1955).

[4] Blakley, R. L., *Biochim. Biophys. Acta*, **23**, 654 (1957).

[5] Huennekens, F. M., Osborn, M. J., and Whitely, A. R., *Science*, **128**, 120 (1958).

[6] Vohra, P., Lantz, F. H., and Kratzer, F. H., *J. Biol. Chem.*, **221**, 501 (1956).

[7] Vohra, P., Lantz, F. H., and Kratzer, F. H., *Arch. Biochem. Biophys*, **76**, 180 (1958).

[8] Frisell, W. R., and Mackenzie, C. G., "Methods of Biochemical Analysis", **6**, 63 (1958).

[9] Fantes, K. H., Page, J. E., Parker, L. F. J., and Lester Smith, E., *Proc. Roy. Soc.*, **136**, 592 (1949).

A Pitfall in High Specific Activity Tracer Studies

IN experiments on the effects of radiation on the process of deoxyribonucleic acid synthesis in mammalian cells it was noted that, in some systems, apparent 'radiosensitivity' of the synthetic process varied with the amount of isotopic 'label' administered.

Ehrlich ascites cells were washed and resuspended in a medium of 50 per cent supplemented Hank's solution and 50 per cent human serum (10^7 cells per ml. medium). Half the culture vessels served as controls and the other half received 2,000 rads of 250 kV. X-rays. Immediately after radiation, tritium-labelled thymidine was added to the vessels as follows: (1) one half of each group, control and irradiated, received 0·5 µc./ml. of specific activity 360 µc./µmole; (2) the other half 5·0 µc./ml. of specific activity 36 µc./µmole, that is, the total amount of thymidine, was increased from 10^{-3} µmole in (1) to 10^{-1} µmole in (2) per culture vessel. All the vessels were then incubated at 37° C. for 2 hr. At the end of the incubation period smears were made for high-resolution autoradiography and grain counting over 500 labelled cells; alternatively, planchettes were prepared for counting total radioactivity of washed cells[1,2]. All experiments were performed with quadruplicate samples.

Under the above conditions, a regular depression of the average grain count or total radioactivity of the cells was noted in the first group, the depression being 50–53 per cent of the control value, while in the second group, the depression was rarely significant, the maximum depression being 12 per cent of the control value.

It was concluded, therefore, that irradiated Ehrlich ascites cells may release deoxyribonucleic acid precursors which are capable of diluting the added label. The dilution to 50 per cent of 10^{-3} µmole of tritium-labelled thymidine indicated a release of compounds equivalent to 10^{-3} µmole of thymidine: if the released compound was thymidine, this would correspond to about 1–2 per cent of the deoxyribonucleic acid thymidine content of 10^7 cells.

No such pool dilution was noted with human bone marrow cells *in vitro*, nor (in preliminary experiments) with mouse thymus cells *in vivo*. These investigations caution workers against the indiscriminate use of 'true tracer amounts' of labelled materials, against the use of high specific activity thymidine in particular, unless previous studies have established the absence of a pool dilution effect.

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[1] Lajtha, L. G., *J. Phot. Sci.*, **2**, 130 (1954).

[2] Lajtha, L. G., Oliver, R., Kumatori, T., and Ellis, F., *Radiation Res.*, **8**, 1 (1958).

BIOLOGY

Control of African Wild Animals

IN the past the control of wild animals, where these conflicted with human interests, has been through destruction, and the rate of destruction has increased greatly in recent years. The possibility of the virtual elimination of the wild ungulates from Africa is a matter for concern if only on the following grounds: (1) The value of these animals as providers of meat under a system of game management and selective cropping. (2) The as yet unassessed effect of the disruption of the natural balance on soil and flora, especially with regard to erosion, desiccation and increase in bush density. (3) The danger of extinction of species and sub-species. (4) The value of these animals as a tourist attraction.

We have become interested in developing effective techniques for transporting animals to areas where the conflict between man and animal is less acute, and where their chances of survival are greater. Prolonged attempts over the years to catch adult animals in appreciable numbers by driving, netting and trapping have failed.

Drugs have been used successfully for catching wild deer in the United States[1] for the purposes of study. This technique has been extended to African species to mark them in order to study changes in population, growth-rates and movement-patterns. Considerable success in this direction has led us to use the method as a preliminary stage in pilot experiments for moving animals to other areas. This work is creating much interest in other parts of Africa and abroad, and demands for information have induced us to publish this early report.

An immobilizing drug is delivered to the animal by a variety of means, and injected intramuscularly. Following capture a tranquillizing drug is then administered before the effects of the first drug have worn off. Transport of the animal then follows.

Most of our results have been obtained using succinyl choline chloride as the immobilizing agent, and Table 1 shows the doses employed.

Table 1. EFFECTIVE DOSES OF SUCCINYL CHOLINE CHLORIDE

Species	Mgm./kgm body-weight Male	Female	No of animals effectively immobilized
Adenota kob thomasi	0·35	0·35	50 (approx.)
Giraffa camelopardalis rothschildi	0·20	0·17	8 (in two doses given at 30 min intervals)
Equus grevyi	0·19	—	2
Kobus defassa ugandae	0·35	—	8
Alcelaphus buselaphus lelwel	0·30	—	1
Diceros bicornis bicornis	—	0·13	1 (must be given in neck region)

Small-scale trials have been made in transporting kob (antelope) from the Lugari area, where the sole remaining herd of Kenya kob now exists, and black rhinoceros from the Makindu area, where land is to be reclaimed from bush. Very shortly we hope to make a similar pilot experiment to move some of the few remaining Rothschild giraffe (only about 200 of which are in existence in the world) out of the farming area where they now reside to marginal grazing land in West Suk.

Much more work is required before this method can be applied on a large scale. As with most drugs producing paralysis, the margin between the effective and lethal doses is small. We are investigating a wide range of drugs in order to determine the most suitable for a given species. With improved techniques in the delivery of drugs and their administration, new opportunities for scientific research and practical wild life management will develop. Although killing of animals must continue under systems of cropping and a degree of control work by shooting will continue to be necessary, it is hoped that the methods of immobilizing and transporting animals that are being evolved will contribute materially to the preservation and proper use of wild life in Africa.

We are indebted to the members of the Uganda and Kenya Game Departments for their co-operation and provision of equipment, Palmer Chemical and Equipment Co., Ltd., for gifts of equipment, and Messrs. Burroughs Wellcome, Ltd. (East Africa), for donation of the succinyl choline chloride.

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[1] Crockford, J. A., Hayes, F. A., Jenkins, J. H., and Feurt, S. D., Trans. North Amer. Wildlife Con., 22, 579 (1957).

Inactivation of Acid Phosphatase in Human Prostatic Homogenates *In vitro*: Role of Calcium

IN the course of metabolic experiments carried out on homogenates prepared from fresh human adenomatous prostates, we have observed that incubation of such homogenates at 37° C. and *p*H 7 in *tris* buffer containing calcium chloride (0·02 *M*) resulted in a rapid loss of phosphatase activity averaging 44 per cent after 3 hr. in 32 experiments. When adenosine triphosphate (0·015 *M*) was added at the beginning of the incubation period, the loss of activity was reduced to an average value of 5 per cent under the same experimental conditions. Addition of adenosine triphosphate during or at the end of the incubation period resulted in complete regeneration of the acid phosphatase activity on further incubation (Fig. 1). We have observed also that adenosine triphosphate could be replaced by sodium diphosphate or ethylenediamine tetraacetic acid (versene) with identical results.

Loss of acid phosphatase activity did not occur or was minimal in prostatic homogenates prepared without the addition of calcium chloride.

These experiments indicate that calcium ions reversibly inactivate the acid phosphatase activity of incubated human prostatic homogenates. Reversal of the process occurred when calcium ions were removed by the addition of phosphate or other

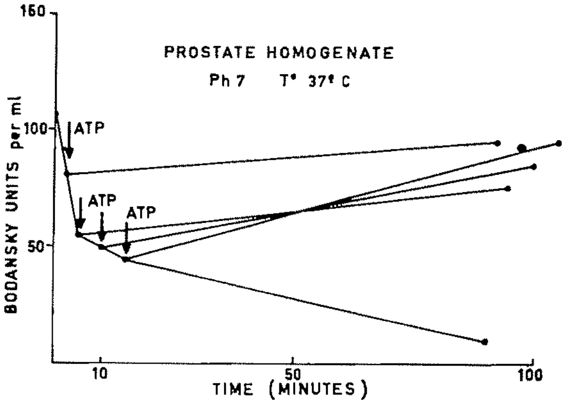

Fig. 1. Effect of adenosine triphosphate on inactivation of acid phosphatase

calcium-binding compounds. This fact, which apparently has not been described up to the present, should be taken into account in the evaluation of fluctuations of acid phosphatase in human serum, whether these fluctuations are spontaneous or associated with certain forms of therapy.

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Influence of Ejaculation Frequency on the Time required for Sperm Formation and Epididymal Passage in the Bull

RECENT experiments have shown that in bulls injected with radioactive phosphate (phosphorus-32), 48–50 days elapse from the moment of injection to the time when radioactive phosphorus appears in the deoxyribonucleic acid of ejaculated spermatozoa[1,2]. This is the period of time required for the formation of spermatozoa in the testes and their passage through the epididymides. The purpose of the present study was to ascertain whether and to what extent this period can be influenced by varying the frequency of ejaculations.

Three bulls ('18', *F* and *Ph*) were used for the experiments, all of the Red Danish milk breed. Labelled phosphate was administered intramuscularly, the dose varying from 5 to 20 μc. phosphorus-32 per kgm. body-weight. Semen was collected by means of an artificial vagina. Throughout the experimental periods as well as during the extended periods preceding and following each experiment the semen was examined for sperm concentration, motility, and the presence of abnormal forms. No deviation from the normal semen picture was encountered in any of the injected animals. The fraction containing deoxyribonucleic acid was separated from the ejaculated spermatozoa using a modification of the Schmidt–Thannhauser method[3] and analysed for phosphorus content and radioactivity. The experimental periods were of 100–130 days duration.

The first series of experiments was carried out on bull '18' which in the previously reported investigation[1] was used at a rate of two ejaculations a week This bull was injected with phosphorus-32 on November 20, 1957. During a period of 45 days prior to the injection no ejaculates had been collected, thus allowing ample time for the establishment of a proper reserve of sperm in the epididymides. Single ejaculates were collected on days 13, 43, 47 and 50 after the injection. The first labelled deoxyribonucleic acid was encountered in the spermatozoa ejaculated on the fiftieth day. Then, on May 1, 1958, the same bull was injected again. However, during a period of 40 days prior to this injection the bull had been subjected to very frequent semen collections so as to deplete the epididymal store as much as possible. Maximal sperm output was also maintained during the actual experimental period by collecting the semen almost every day. Nevertheless, the first semen sample to contain sperm with labelled deoxyribonucleic acid was that obtained 48 days after the injection of radioactive phosphate.

The second series of experiments was carried out on bulls *F* and *Ph*, mainly to find out if age of the animal or season of the year can influence the time required for sperm formation and maturation.

For a period of 1½ months preceding the actual experiments, bulls *F* and *Ph* had been subjected to collections at a rate of 2–3 a day and 1 a week, respectively. Phosphorus-32 was injected to both bulls on November 5, 1958, and then again 76 days later. The rate of collections during the experimental period of 105 days after the first injection was the same as during the pre-experimental period. On the one hundred and sixth day after the first injection, that is, on the thirtieth day after the second injection, the collection procedures were reversed, that is, from now onwards bull *Ph* was used for collection 2–3 times daily, while bull *F* was used only once a week. Moreover, just prior to the time when this reversal was instituted the sperm reserve of *Ph* was quickly depleted by performing on the animal an exhaustion test in the form of 14 rapidly succeeding collections. Table 1 illustrates the ejaculation schedule followed for the bulls *F* and *Ph*.

Table 1. EJACULATION SCHEME FOR THE BULLS *F* AND *Ph*

Bull (No.)	Injection of phosphorus-32	Rate of ejaculations during periods corresponding in length to :	
		Spermatogenesis	Epididymal passage of sperm
F	first	2–3 daily	2–3 daily
	second	2–3 daily	1 per week
Ph	first	1 per week	1 per week
	second	1 per week	2–3 daily

The aims of planning the experiment in the way outlined above were as follows. Constant frequency of collection was maintained during periods corresponding in length to the time necessary for the completion of spermatogenesis since one could scarcely expect the spermatogenesis, as distinct from sperm passage through the epididymis, to be affected by the number of spermatozoa withdrawn from the animal by means of semen collections. During the remaining periods, however, which would correspond in length to the time required for the passage of sperm, the frequencies were varied deliberately. Thus if varying ejaculation frequencies could affect the epididymal passage of spermatozoa at all, the appearance of labelled deoxyribonucleic acid in ejaculated sperm would occur at different times from the moment of injection according to the respective rates of ejaculations. In rams[4] and rabbits (Koefoed-Johnsen, H. H., and Foote, R. H., unpublished results) it has been previously found that labelled spermatozoa were present in the head of the epididymis about 30 days after the administration of the radioisotope. It was thought reasonable to apply to bulls a similar figure although, admittedly, this period could be possibly longer, but in any event not shorter, in the bull.

All results obtained on the three bulls are given in Table 2. This table shows the time interval between the phosphorus-32 injection and the first appearance in ejaculated semen of spermatozoa containing labelled deoxyribonucleic acid. It indicates also the average age of spermatozoa in the ejaculates as assessed by the time required by ejaculated spermatozoa to reach the maximum of labelling (average

Table 2. APPEARANCE OF RADIOACTIVE PHOSPHORUS IN THE DEOXYRIBONUCLEIC ACID (DNA) OF EJACULATED SPERMATOZOA AFTER INTRAMUSCULAR INJECTION OF LABELLED INORGANIC PHOSPHATE TO BULLS

Bull (No.)	Age (months)	Date of phosphorus-32 injection	Dose of phosphorus-32 (μc./kgm.)	No. of ejaculates		Number of sperm ejaculated $\times 10^9$			Time interval (days) from injection to .	
				During days 0–30	From the thirtieth day onwards and until the appearance of labelled DNA	From injection to the appearance of labelled DNA	During days 0–30	From the thirtieth day onwards and until the appearance of labelled DNA	First appearance of phosphorus-32 in DNA	Labelling of DNA reaching the maximum (average)
18	30	May 1, 1957	11	22	14	104·4	65·9	38·5	48	60
18	36	Nov. 20, 1957	20	1	3	38·6	15·1	23·5	50	69
18	42	May 1, 1958	12	49	37	186·7	132·9	53·7	48	52
F	12	May 17, 1958	5	22	—	39·5	24·4	15·2	48	—
F	18	Nov. 5, 1958	8	71	50	122·9	77·8	45·1	47	55
F	21	Jan. 20, 1959	7	79	5	93·0	60·1	32·9	50	66
Ph	30	Nov. 5, 1958	8	5	6	79·9	29·7	50·2	51	63
Ph	33	Jan. 20, 1959	10	5	67	244·5	76·7	167·7	49	58

values). Table 2 includes also results for the number of ejaculates obtained during definite periods of time and the number of spermatozoa ejaculated during the same periods. The general conclusion which can be drawn from these results is that the period of time required for the appearance in ejaculates of spermatozoa with labelled deoxyribonucleic acid is remarkably constant irrespective of the frequencies of ejaculations. The average age of ejaculated spermatozoa, however, varies according to the frequency of collections; thus, infrequent collections will result in a higher average age of the spermatozoa in ejaculated semen, indicating that the sperm have been stored in the cauda epididymidis for a longer period of time than in the case of frequent ejaculations.

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[1] Dawson, R. M. C., *Nature*, 181, 1014 (1958).
[2] Koefoed-Johnsen, H. H., Roy. Vet. and Agric. Coll., Sterility Res. Inst. Ann. Rep., 18 (Copenhagen, 1958).
[3] Schmidt, G., and Thannhauser, S. J., *J. Biol. Chem.*, 161, 83 (1945).
[4] Ortavant, R., *C.R. Soc. Biol., Paris*, 148, 804 (1954).

Abnormalities in the Urinogenital System of the Common Dogfish

ABNORMALITIES of the urinogenital system of the common dogfish (*Scyliorhinus caniculus*) have hitherto been regarded as infrequent[1–3]. It is therefore of interest to note that in the past academic year three abnormal dogfish have been found by this department.

Case 1. This dogfish was 14·3 in. long (36·3 cm.) from the tip of the snout to the posterior end of the first dorsal fin. The external appearance was that of a young male. The right clasper was 3·5 cm. long, and the left clasper 2·9 cm. long. The united mesial border of the pelvic fins[3] was notched to a depth of 9 mm. So far as can be judged, the structure of the siphon was normal. The appearance of the mouth[3] was typically male.

Internally the urinogenital ducts were symmetrical and hermaphrodite. Superficially, the urinary and Wolffian ducts presented the normal appearance of a young male, and the seminal vesicles were dilated to a diameter of 7 mm. There was, however, no sperm sac. The main urinary duct on each side opened into the urinary papilla, but the seminal vesicles opened into the dorsal wall of the base of each oviduct.

On each side the Müllerian duct extended from the common oviducal aperture to the cloaca. The length of the common oviducal aperture was 9 mm. There was no indication of an oviducal gland. Throughout the major part of its length the diameter of the Müllerian duct was 2·5 mm., and the lumen was filled with a clear mucus. The posterior part of the Müllerian duct had an average diameter of 5 mm., so this part was apparently dilating, as would be expected in a young female prior to copulation. The right Müllerian duct was closed at the cloaca, and sealed to the lateral side of the urinary papilla. The equivalent region of the left side was damaged by student dissection, but is believed to have been in the same condition originally.

The left gonad had the external appearance of a functional testis. Microscopical examination revealed that proliferation of spermatogonia was occurring in the seminiferous tubules, but that maturation of spermatozoa was not occurring. The right gonad was less well developed, and had the appearance of an immature ovary. Oocytes as big as 2 mm. in diameter were visible in one part.

Case 2. The dorsal fin had been removed with the tail, but the animal was 13 in. long (33 cm.) from the tip of the snout to the posterior end of the pelvic fin. In other words the animal was slightly longer than case 1. The external appearance was not so typically male. The left clasper only was present, and was only 2 cm. long. The pelvic fins were completely separated posteriorly as in a typical female. There was no indication of a siphon having been present.

Internally, as in case 1, the urinogenital ducts were bilaterally hermaphrodite, and the sperm sac was absent. Again the urinary ducts presented a typical male appearance; but in this case they opened into the dorsal wall of the oviduct, and no urinary papilla was present. The seminal vesicles were indicated, but were only 3 mm. in diameter. As in case 1 they opened into the dorsal wall of the base of the oviduct. The Müllerian ducts extended the whole length of the body cavity, with an average diameter of 2 mm., and dilating in the oviducal gland region to 6 mm.

The left gonad had the external appearance of an abnormally short testis. The right gonad had the appearance of an immature ovary.

Case 3. A new pectoral steak was found to contain both vasa deferentia and an immature ovary with oocytes. The rest of the animal was not located.

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[1] Bamber, Ruth C., *Proc. Zool. Soc. Lond.*, 16, 217 (1917).
[2] Arthur, D. R., *Proc Linn. Soc. Lond.*, 162, 52 (1950).
[3] Chapman, G., *Proc. Zool. Soc. Lond.*, 121, 511 (1951).

Formation of New Wool Follicles in the Adult Sheep

It has generally been held that the formation of new hair follicles from the epidermis does not occur after the adult complement has been established. Evidence to the contrary was published by Ribbert[1] as early as 1904, and more recent work in this field has confirmed that new hair follicles can in fact develop from adult epidermis[2-5]. This phenomenon has not, to our knowledge, been reported in the sheep and the present observations, describing the formation of new wool follicles in adult animals, were made by chance during the examination of unsuccessful homografts.

Pieces of skin (about 3 mm. × 3 mm.) from 49–80 day Merino sheep fœtuses were grafted on to the abdominal walls of their mothers, the graft beds having been prepared by the removal of pieces of full-thickness maternal skin. When these sites were biopsied, 15–62 days after grafting, and examined microscopically, the presence of immature follicles was noticed. Neither the age of fœtal skin when grafted, nor the time that elapsed between application of the grafts and sampling of the site, seemed to have influenced the occurrence of new follicles. They were considered to be maternal and not fœtal in origin, due to the presence of features which could only be associated with adult skin, for example: (*a*) mature follicles, including bundles of secondaries; (*b*) shedding and regenerating follicles; and (*c*) fibre remains in the dermis.

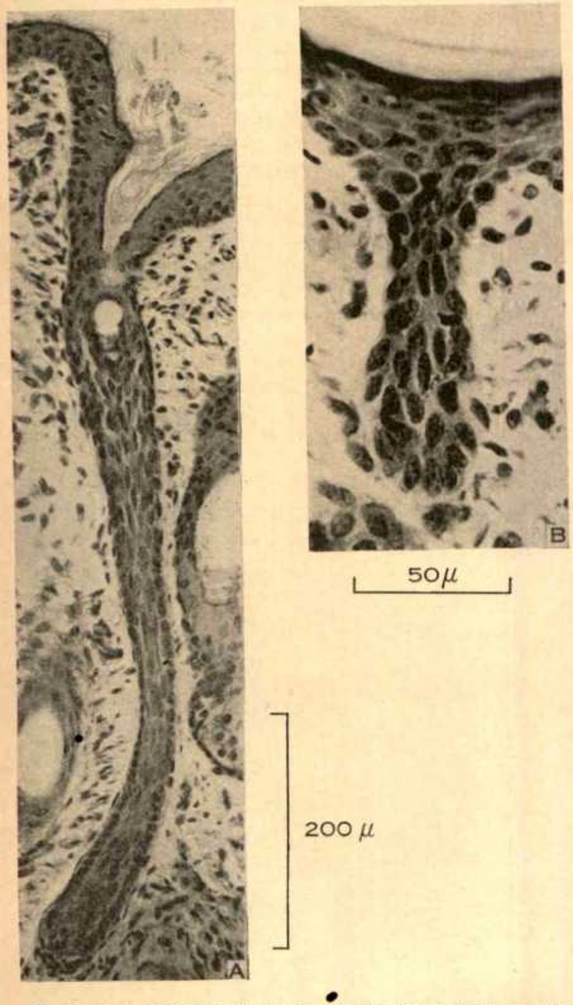

Fig. 1. Longitudinal sections of new original wool follicles on the abdomen of an adult sheep. *A*, New follicle with a dermal papilla; *B*, new follicle at an earlier stage without dermal papilla cells

Both regenerating and new wool follicles were observed. The former developed from below the sebaceous gland-level of a resting follicle and were associated with original dermal papillæ. This process in sheep has been briefly described by Burns[6]. New follicles were of two kinds, derived and original; derived were formed from the neck region of existing follicles, and original from the epidermis, in a manner similar to that described in the sheep fœtus[7]. Their papillæ also, if present, were newly formed.

There was considerable variation in the stages of development reached by both original and derived new follicles. In their early forms they appeared as slender epidermal downgrowths, generally, though not invariably, associated with dermal papilla cells (Fig. 1*A* and *B*). Sebaceous glands and hair canals had been formed in the more advanced follicles, but no sweat glands were observed. The most advanced follicle contained a fibre about to emerge above the skin surface. However, since the majority of the new follicles were in early stages of development (prior to the formation of the hair cone), it cannot certainly be claimed that this neogenesis would have culminated in the general emergence of new fibres.

Breedis[2] found that the prevention of wound contracture was a pre-requisite for the formation of new hair follicles, and it seems reasonable to suppose that in the present experiments this was achieved by the presence of the fœtal homografts. The possibility that an interaction between fœtal and adult tissue was responsible for the phenomenon of neogenesis cannot be dismissed. However, in the absence of distinguishable fœtal skin, our findings show that the skin of adult sheep has not entirely lost the potential for the initiation of new wool follicles.

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[1] Ribbert, H., *Arch. Entw. Mech. Org.*, **18**, 578 (1904).
[2] Breedis, C., *Cancer Res.*, **14**, 575 (1954).
[3] Billingham, R. E., and Russell, P. S., *Nature*, **177**, 791 (1956).
[4] Kligman, A. M., and Strauss, J. S., *J. Invest. Derm.*, **27**, 19 (1956).
[5] Billingham, R. E., in "The Biology of Hair Growth", ed. Montagna, W., and Ellis, R. A., 451 (Acad. Press, New York, 1958).
[6] Burns, M., *J. Agric. Sci.*, **39**, 64 (1949).
[7] Hardy, M. H., and Lyne, A. G., *Aust. J. Biol. Sci.*, **9**, 423 (1956).

Penetration of *Fasciola gigantica* Cobbold, 1856 into Snail Hosts

I have recently demonstrated[1] that penetration of *Fasciola hepatica* into *Limnaea truncatula* is a brief but elaborate process which involves the successful attachment of the miracidium to the snail, seemingly by suctorial adhesion, and the production of a secretion which cytolyses the integument of the snail in a circumscribed area underlying the anterior non-ciliated pit of the larva. Not until the snail's

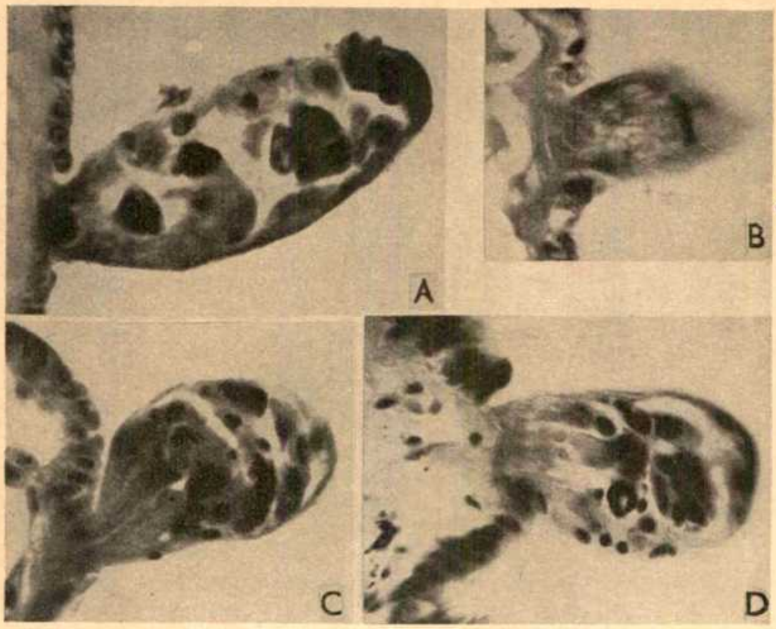

Fig. 1. *Fasciola gigantica* sections (8μ thick) showing early stages of penetration. *A* and *B*, miracidium attached to mantle (anterior end only in *B*). *C* and *D*, miracidium attached to body

epithelium has been perforated by cytolytic action does the larval trematode thrust itself into the snail's body. By the time these preparations for entry have been completed, the miracidium has cast off its ciliated epithelium, so that although it is the miracidium which attacks the snail, it is the young sporocyst which enters the snail's body.

It is now possible to demonstrate that the process is precisely similar during penetration of *Fasciola gigantica* into certain snails, all three species of which became infected, though not with equal facility or intensity. Normal routine procedures were followed in establishing infection, and periods of 30, 60 and 120 min. which were allowed for penetration were adequate. It is necessary to state further that the serial sections and photomicrographs here referred to concern snails which are believed to belong to the species *Limnaea rufescens* Gray, which has been once regarded[2] as merely a race of the well-known snail *Limnaea auricularia*, and was designated "*L. auricularia rufescens*" in a letter published in these columns[3]. What is stated, however, is true also for *Limnaea natalensis*, and probably true for *Limnaea acuminata*, although penetration into this species was seldom seen.

During early preparations for penetration which the miracidium of *Fasciola gigantica* makes when attached to the mantle epithelium, the anterior papilla of the larva is retracted in what must be regarded as suctorial action (Fig. 1*A*). Even when the larva has made contact with the sub-epithelial tissues of the snail, the anterior extremity of the larva is remarkably obtuse and could not look less like a piercing organ (Fig. 1*B*). This is true also for sections which reveal the extent of the damage done to columnar epithelia (Fig. 1*C* and *D*). As is seen during penetration of *Limnaea truncatula* by *Fasciola hepatica*, the damaged epithelial cells of the foot or body which protrude at the rim of the perforation created by cytolysis tend to become heaped externally to form a chimney-like eminence around the anterior region of the larva. At this stage the miracidium likewise casts off the ciliated cells of the most anterior tier (Fig. 2*A*). Soon afterwards, the larva is pushing this denuded anterior region of the body (as well as the anterior papilla) into the opening (Fig. 2*B*). The more posterior ciliated cells of the larva are then becoming loosened from underlying tissue, and ultimately they are cast off as well. When still later the larva squeezes through the opening into the body of the snail (Fig. 2*C*) it is evidently devoid of a ciliated investment, so that without a doubt the entering larva is a globular young sporocyst (Fig. 2*D*).

The cytolytic secretion which is produced by the miracidium of *F. gigantica*, like that produced by the larva of *F. hepatica*, may be termed 'digestive' in the sense that the larval gut is concerned, as are paired gland cells, in its production. In both species of miracidia the gut comprises a syncytium having a typical cluster of four nuclei, and both a mouth and a lumen can be seen in some sections. The secretion is acting on the snail's epithelium at a time when the miracidium is in fact undergoing metamorphosis. In both species therefore it is a young sporocyst which enters the snail, although it is the miracidium which

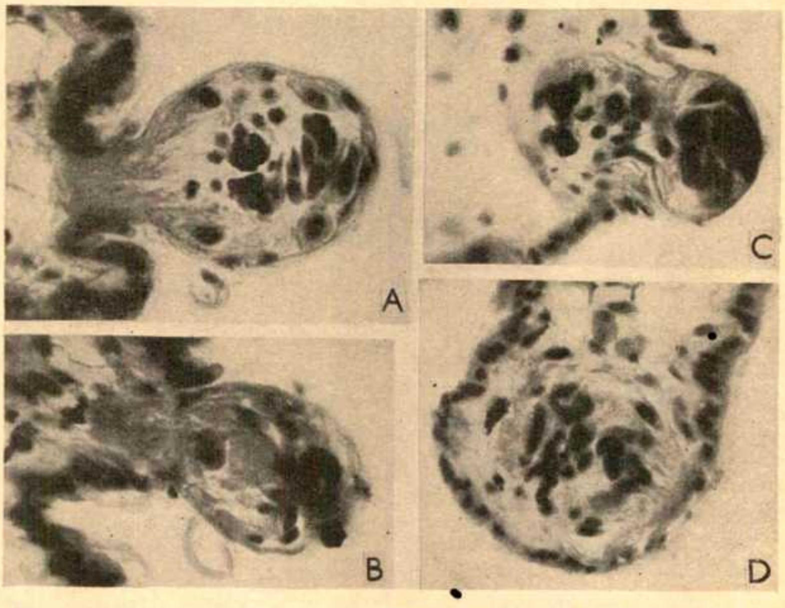

Fig. 2. *Fasciola gigantica* sections (8μ thick) showing later stages of penetration. *A* and *B*, miracidium casting off ciliated cells. Note broken cells heaped at margin of opening. *C*, partial entry of young sporocyst. *D*, complete entry

first adheres to the snail's integument and then cytolyses it to the ultimate perforation. Observations at present being made indicate that penetration of *Schistosoma mansoni* into *Australorbis glabratus* is a closely similar process, giving further reason to believe that entry into the molluscan host is likely to be a unified process of the kind indicated here in all the digenetic trematodes which have a ciliated miracidium.

My thanks are due to Mr A. T. Green for taking the photographs.

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[1] Dawes, Ben, *Nature*, **184**, 1334 (1959).

[2] Hubendick, B., *K. Svenska Vetenskapsakad. Handl.*, **3**, 1 (1951).

[3] Kendall, S. B., and Parfitt, J. W., *Nature*, **171**, 1164 (1953).

Association-Analysis in Plant Communities

THE problem of subdivision of a population characterized by a correlation matrix obtained from a set of 2 × 2 contingency tables has recently been the subject of communications by Williams and his colleagues[1,2]. The basis of Williams's method is to choose that species which, by its presence or absence, subdivides the population of ecological observations in the most efficient manner possible. The purpose of this communication is to question the advisability of placing so much reliance on a single species, no matter how well selected.

Following Goodall's[3] discussion we may define a homogeneous unit of vegetation as one in which all species associations are indeterminate or non-significant. Two such units may then be specified as:

	Unit 1	Unit 2
Species *A*	a_1	a_2
Species *B*, etc.	b_1	b_2

where a_1 is a value of unity or less representing the chances of species *A* being present in a quadrat of specified size within a stand of unit 1, the chances of joint occurrence of species *A* and *B* in quadrats of that unit being given by a_1b_1. With a large population of quadrat samples composed of equal numbers of the two units the only subdivision which will reduce all residual associations to non-significance at the first dichotomy is that which reproduces the two units: and this division will be attained by Williams's method when one species is universally present in one unit and universally absent in the other. If the two units are of very different floristic structure many species may fulfil these requirements, and the division will be obvious without elaborate calculation. Conversely, it is probable that when computational assistance is most needed on account of the resemblance of the two units, and most likely to be employed, no species will be so conveniently distributed. It may then be necessary to rely for division on species *X*, for example, with values of $x_1 = 0{\cdot}9$, $x_2 = 0{\cdot}1$, which would misclassify 10 per cent of the quadrats and produce groups in which association had not been reduced to non-significance. Further subdivision and recombination may yield a very much closer approximation to the two constituent units, but recombination can be a questionable process[2].

In this simple theoretical example the actual species (X) selected for subdivision of the population by Williams's method depends on the values of x_1 and x_2, since (apart from an unlikely circumstance) the species with the greatest $\Sigma|r|$ corresponds to the one with the highest value of:

$$\frac{|x_1 - x_2|}{\sqrt{(x_1 + x_2)(2 - x_1 - x_2)}} \text{ or } f(X)$$

The greatest possible value of this function is 1 when $x_1 = 1$, $x_2 = 0$, so that as has already been suggested a species universally present in one unit and universally absent in the other is automatically selected if such exists. In the absence of any species approaching this ideal, $\Sigma|r|$ might choose species Q having $q_1 = 0{\cdot}4$, $q_2 = 0{\cdot}0$ ($f(Q) = 0{\cdot}5$), which misclassifies 30 per cent of the sites. This species would be chosen in preference to P which, with $p_1 = 0{\cdot}74$, $p_2 = 0{\cdot}26$ ($f(P) = 0{\cdot}48$), only misclassifies 26 per cent of the sites. Whereas one of the groups sorted out by Q has no significant association, the total residual level must in some sense be higher than that left by division on P, so that the species selected by $\Sigma|r|$ (or $\Sigma\chi^2$ since in this case the two give the same answer) will not necessarily be that one which subdivides the population in the most efficient manner possible. "Theoretical considerations, it will be recalled, tell us that it is *likely* to do so, not that it will"[2].

Goodall[4] has also advocated the use of factor analysis methods in ecological work, and it is tempting to apply them to a matrix built up from presence and absence data. But these methods are intended for use with continuous variables and are not necessarily suited to such a matrix: in fact the scatter of the first two factors may be misleadingly distorted (Harberd, D. J., unpublished work). Furthermore, extraction of more than two factors only yields a confusing array of statistics very difficult to visualize and interpret. However, with presence and absence data the distance between two sites in multi-dimensional space (an approximation to Mahalanobis's D^2) can be taken as the number of species found once only in the two species lists. This quantity, hereafter called d^2, is easily determined by inspection of species lists and has several attractive properties. Thus, so long as the relevant part of the species log area curve is linear, its value between stands of the same vegetational unit is independent of the quadrat size used, and characteristic of that unit.

A complete table of d^2 between the sites in a particular study is easily though laboriously constructed. One such table relating to 80 local bent-fescue sites was fairly easily sorted by eye into seven less heterogeneous groups characterized by low mean d^2 within groups relative to that between groups.

Table 1. CHANCES OF FINDING SOME SELECTED SPECIES IN TURVES CUT WITH SAMPLING TOOL IN EACH OF SEVEN SITE GROUPS

Species	Site groups						
	1	2	3	4	5	6	7
Agrostis stolonifera	0·37	0·06	—	—	—	—	—
Sagina procumbens	0·66	0·35	0·14	0·12	—	—	—
Cardamine pratensis	0·60	0·55	0·22	0·05	0·11	—	—
Ranunculus repens	0·54	0·31	0·22	0·10	0·01	—	0·01
Poa trivialis	0·77	0·49	0·22	0·15	0·06	—	0·02
Trifolium repens	0·97	0·91	0·70	0·82	0·74	0·60	0·17
Festuca rubra	1·00	1·00	1·00	0·97	0·94	0·77	0·54
Cerastium vulgatum	0·14	0·58	0·38	0·38	0·31	0·10	0·02
Cynosurus cristatus	0·29	0·29	0·44	0·35	0·19	0·03	—
Galium pumilum	0·03	0·09	0·06	0·20	0·03	—	—
Prunella vulgaris	—	0·24	0·12	0·37	0·31	0·10	—
Carex caryophyllea	—	0·04	0·22	0·53	0·74	0·50	0·12
Viola riviniana	—	0·02	0·02	0·18	0·44	0·17	0·01
Luzula campestris	—	0·06	0·28	0·57	0·51	0·90	0·50
Festuca ovina	—	0·06	0·44	0·57	0·87	0·87	0·95
Galium hercynicum	—	—	—	0·07	0·03	0·30	0·65

(The possibility of using a computer for this process is at present being investigated.) Furthermore, the groups fell into a series (numbered 1–7 from flushed to leached), the mean between group d^2 being lower for neighbouring groups than for more distant groups. Reconstructing the floristic composition of the groups from the species lists, it can be seen that each species tends to attain its greatest frequency in one of the groups, tailing away on either side (Table 1). In no case in the whole table is a species universally present in one site group and universally absent in the next: we are, in fact, considering the case postulated of closely related vegetational units where differences are in relative frequency rather than in presence and absence. The species with the highest $\Sigma\chi^2$ is *Festuca ovina*, which increases gradually in frequency across the whole breadth of the table.

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[1] Williams, W. T., and Lance, G. N., *Nature*, **182**, 1755 (1958).
[2] Williams, W. T., and Lambert, J. M., *J. Ecol.*, **47**, 83 (1959).
[3] Goodall, D. W., *Aust. J. Bot.*, **1**, 39 (1953).
[4] Goodall, D. W., *Aust. J. Bot.*, **2**, 304 (1954).

PATHOLOGY

Histotrophic (Parasitic) Stages of *Hæmonchus contortus*

Work on resistance and immunity to parasitic infections over the past thirty years has tended to support the view that the immune response, which has been shown to be humoral in nature, is stimulated by the excretions and secretions of the invading parasite[1,2].

It was first reported by Stoll[3] that sheep, after undergoing an initial infection, develop a resistance to reinfection with *Hæmonchus contortus*. Later, Stoll[4] reported that he was able to induce resistance by injecting, intraperitoneally or subcutaneously, sterilized exsheathed infective *H. contortus* larvæ.

At this Institute attempts to repeat this work met with failure; however, it was later learned[5] that the immunizing doses used by Stoll (4,000–22,000 larvæ per kgm. body-weight) were far greater than the numbers of larvæ used for inoculations in our unsuccessful experiments[5].

Following the unsuccessful vaccine trials a series of experiments were designed to elucidate further the mechanism of resistance to *H. contortus* and if possible to determine the source of antigen and site of the antibody response.

Three groups of sheep were used: (1) worm-free lambs; (2) worm-free sheep (one year old); (3) resistant sheep (one year old). Animals from each group were challenged with oral doses of 5,000–100,000 *H. contortus* larvæ and killed at regular intervals during the second to fifteenth day after infection. After slaughter, the abomasum was quickly opened and treated in the following manner: (*a*) Pieces of the pyloric and cardiac portions were cut out and fixed for histological examination. (*b*) The mucosa was then scraped off into warm saline. Some of the histotrophic stages were quickly collected, rewashed in warm sterile Tyrode and transferred into previously prepared sera from susceptible and resistant sheep. (*c*) Another sample of histotrophic larvæ was collected and pooled with similar stages from other sheep for use in protection tests. Such larvæ were either directly injected into susceptible sheep or deep-frozen for later use. (*d*) The remaining mucosal scrapings were fixed in formalin for detailed morphological studies of the parasitic stages.

The results of these studies may be briefly summarized as follows:

Morphology. Morphological studies of the histotrophic stages revealed that the rate of development of the parasitic phase is dependent upon the age and immunological state of the host. In susceptible lambs most infective (that is, L_3 stage) larvæ have undergone differentiation and completion of the first parasitic ecdysis by the third day after infection (that is, to the L_4 stage) but no marked growth has occurred. The larva begins to increase in size, reaching 1–1·5 mm. about the fifth day, when a second ecdysis takes place. Sexual differentiation is apparent in the L_5 stage and the young adult reached 2–5 mm. in length by the seventh to ninth day. Eggs are first produced between the twelfth to fifteenth day. In older susceptible sheep the first parasitic ecdysis is not generally completed until the fifth day and the entire cycle is delayed, with eggs first being produced on the sixteenth to twenty-fourth day. In resistant sheep the life-cycle of the parasite proceeds to the L_4 and L_5 stages by the fifth to seventh day, but the majority of worms become inhibited at these stages until they are thrown off, usually by the tenth to fifteenth day.

Histology. Histological examination of the mucosa and the L_3, L_4 and L_5 histotrophic larvæ *in situ* has so far revealed no evidence of host tissue responses to these stages in either susceptible or resistant animals up to the ninth day. Damage to host tissue first occurs after the tenth day, when young adults begin to burrow into the mucosa to suck blood.

The histotrophic larvæ appear to pass their existence within the paramucosal lumen, that is, below the mucous layer on the surface and in the crypts of the villi. In resistant animals where there is evidence of inhibited development of the histotrophic stages, the effect is apparently produced entirely through humoral agencies.

Serology. Studies on the *in vitro* reaction of serum to exsheathed L_3, L_4 and L_5 larvæ and adult worms were carried out using sheep sera which had been serially diluted with phosphate buffer, some with and some without complement. It was found that complement was not required and that precipitations occurred at dilutions up to 1 : 40. A minimum time of 10–12 hr. appeared to be required and precautions had to be taken to produce bacteria-free preparations. Most larvæ remained active for up to 36 hr. of immersion.

The L_4 and L_5 stages showed the greatest antigenic activity, overnight preparations revealed precipitates along the inside of the intestine, a plug formation at the rectal sphincter and precipitated globules at the excretory pore. Neither L_3 larvæ nor adult worms showed any apparent reaction to immune serum although the exsheathed L_3 began to show a form of streaming oral precipitate after about 36 hr. Adult worms remained active for up to 72 hr. and continued to produce eggs for most of this time.

Protectivity. The protection experiments showed that the L_4 and L_5 stages were significantly more antigenic than the L_3 stages. A protection of about 70 per cent (based on abomasal worm counts) was achieved in two groups of sheep, one group of which received injections of several thousand fresh L_4 and L_5 stages and the other group similar quantities of 0·5 per cent formalin-treated L_4 and L_5 larvæ. No protection could be induced below 100,000 with L_3 larvæ and the best that was achieved with larger amounts was a 50 per cent advantage over controls.

The evidence from these studies indicates that the histotrophic stages of *H. contortus* are important sources of antigen and appear to be the antibody-susceptible stages.

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[1] Chandler, A. C , *Rice Inst Pamphl.*, 45, 4 (1958)
[2] Soulsby, E. J. L., *Vet. Rec* , 69, 1129 (1957).
[3] Stoll, N. R , *Amer. J. Hyg* , 10, 384 (1929).
[4] Stoll, N. R , *J. Parasit.*, 28, Supp. 20 (1942).
[5] Stoll, N. R., *Rice Inst. Pamphl* , 45, 184 (1958).

'Carcinolipin': an Endogenous Carcinogenic Substance

A FACTOR affecting protein synthesis has been recently detected in egg-yolks[1]. This substance has since been isolated in crystalline form and it is possible to obtain the same crystalline material from animal tissues also[2]. Liver and other tissues from tumour-bearing rats contained a higher amount of this compound when compared with the same tissues from normal animals[3].

This material enhances the incorporation of labelled amino-acids into proteins of Ehrlich ascites cells and tissue-homogenates *in vitro*[2]. Also the incorporation of labelled amino-acids into proteins of rat liver microsomes is stimulated in the presence of this substance[4], and it seems that it is capable of influencing protein biosynthesis in its every stage[5]. The net-synthesis of serum albumin in rat liver slices *in vitro* is greatly enhanced by the addition of this compound into the incubation mixture[6].

The effect on the incorporation of labelled amino-acids into ascites cells protein appears to be strikingly dependent on dose Some doses have stimulatory activity while others cause an inhibition of this process[2]. The same general pattern was also found when the effect of carcinogenic hydrocarbons was tested in the same system. Related non-carcinogenic hydrocarbons are free of this activity, which seems to be rather specific for carcinogenic substances[7]. For this reason carcinogenic activity was presumed for our substance mentioned above.

To investigate this possibility further, 50 Wistar rats of both sexes, 8 months old, were injected subcutaneously with this compound (3 mgm. each) in olive oil. Control group of 50 animals received the same amount of pure olive oil.

The incidence of tumours in the experimental group is given in Table 1. After fourteen months no tumours have been found in the control group. An unusually high incidence of chronic pneumonia with frequent bronchiectasias (10 cases) appeared in the experimental group (no case in the controls as yet). This disease is known to occur fairly often in old rats[8] but not in younger ones.

This experiment is of course not yet completed, and there are further tumours appearing in the experimental group. However, it is clear already from these preliminary results that our compound has definite carcinogenic activity.

Table 1. TUMOUR INCIDENCE IN RATS TREATED WITH 'CARCINOLIPIN' (Latent period is given in months after single subcutaneous administration of 3 mgm. 'carcinolipin')

Latent period	Site	Histological appearance
5	Subcutaneous (site of injection)	Sarcoma
9*	Abdominal cavity	Carcinoma
9†	Lungs and abdominal cavity	Carcinoma
9	Subcutaneous (site of injection)	Sarcoma
11	Subcutaneous (site of injection)	Sarcoma
12	Mammary gland	Fibroadenoma
13	Mammary gland	Fibroadenoma

* In this case a very large tumour was found originating perhaps in the liver tissue.

† Very large tumours were found in this case, one originating from the lungs, the other being localized in abdominal cavity. Because of the magnitude of these tumours as well as their highly polymorphous appearance, the same in both cases, it was not possible to ascertain the site of the primary tumour.

The chemical nature of this substance is not quite clear. It is of lipid nature and seems to contain a bound phosphorylated pentose, also. Because of the predominantly lipid character of the active substance as well as its carcinogenic properties, the name 'carcinolipin' is proposed for it until its exact chemical structure is made clear.

It is very probable that 'carcinolipin' is responsible for the higher incidence of tumours found in mice fed egg-yolks[9]. It seems quite probable, too, that 'carcinolipin' might be identical with the endogenous carcinogenic substance[10]. A closely similar isolation method followed in our experiments strongly supports this view. Moreover, tumours of various types were found. The ability to produce tumours not only on the site of subcutaneous injection seems to be characteristic of the endogenous carcinogenic substance[11], carcinogenic hydrocarbons, on the other hand, producing almost exclusively subcutaneous sarcomas after this mode of administration.

'Carcinolipin' seems to possess a general role of a growth-promoting factor in tissues and other biological materials, since it stimulates the growth of young rats and chickens when added to their diet (Hradec, J., and Trojan, K., unpublished work). Probably it is capable of inducing malignant growth only in certain circumstances, which are being investigated further in our laboratory.

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[1] Hradec, J , *Nature*, 182, 52 (1958)
[2] Hradec, J , and Štroufová, A , *Biochim Biophys Acta* (in the press).
[3] Hradec, J., Abstracts of Papers, Seventh International Cancer Congress, London (1958).
[4] Hradec, J., *Biochim. Biophys Acta* (in the press)
[5] Loftfield, R B , "Prog. Biophys.", 8, 347 (1957)
[6] Hradec, J., and Dušek, Z , *Biochim Biophys Acta* (in the press)
[7] Hradec, J , *Brit J. Cancer*, 13, 336 (1959).
[8] Griffith, J Q , and Farris, E J , "The Rat in Laboratory Investigations" (Lippincott, Philadelphia, 1942)
[9] Szepsenwol, J , *Proc Soc Exp Biol. Med* , 96, 332 (1957)
[10] Shabad, L M , *Cancer Res* , 5, 405 (1945)
[11] Kleinenberg, H E , Neufach, S A , and Shabad, L M , *Cancer Res* , 1, 853 (1941)

ENTOMOLOGY

Male Genitalia of *Apis* Species

The external parts of the male genitalia of *Apis* species are poorly developed, and an elaborate structural modification of the internal walls of the ædeagus is everted during copulation. Hitherto undescribed except for *A. mellifera*, this apparatus has now been examined in specimens of the other three species from Ceylon kindly obtained by Dr. B. A. Baptist and Mr. L. A. S. Perera.

Fig. 1 shows the organs of *A. indica* and *A. mellifera* (in an over-everted condition which is not reached in natural mating[1]). *A. indica* lacks the hard plates of *A. mellifera* and has the small projections at the bases of the sticky horns enlarged to trilobed structures. The shapes and proportions of the various parts also differ. In the other two species only unverted organs of preserved specimens were examined. Neither has hard plates. *A. dorsata* has three additional pairs of sticky horns, which correspond to the trilobed structures of *A. indica*, and five additional bristly projections. *A. florea* has trilobed structures somewhat like those of *A. indica*, a single pair of sticky horns which are relatively much longer than in the other species, and no bristly projections. The endophallus differs markedly in shape from that of the other species and has fleshy areas (presumably glandular) in its walls.

Endophallic plates, apparently identical with those of European bees, were found in bees from Palestine, Uganda, Tanganyika (received from Dr. F. G. Smith) and South Africa (all of which are considered to belong to sub-species of *A. mellifera*). It would be interesting to know whether they ever occur in the apparently closely allied *A. indica*. Male specimens of both species would be welcomed from regions of possible overlap between them, such as Persia, Afghanistan, Pakistan or northern China. They should preferably be preserved in glycerine.

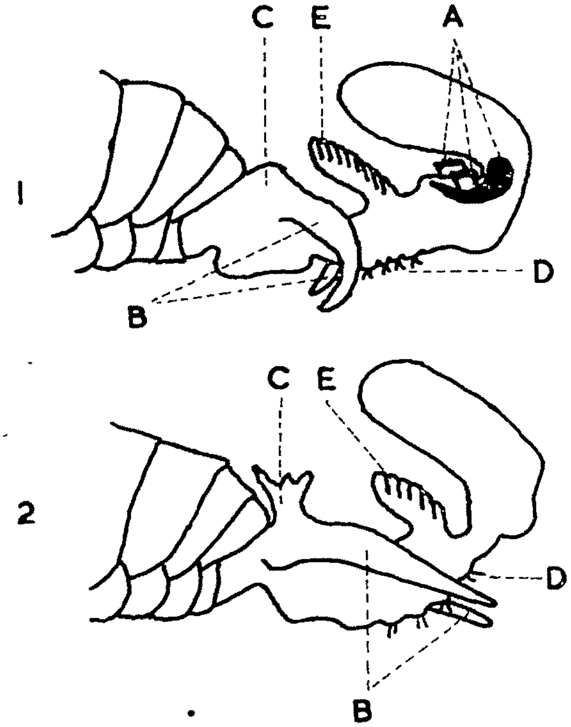

Fig. 1. Male genitalia. 1, *A. mellifera*; 2, *A. indica*. *A*, Hard plates; *B*, sticky horns; *C*, basal projection and homologous trilobed structure; *D*, four hairy projections; *E*, fimbriate lobe

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[1] Woyke, J., and Ruttner, F., *Bee World*, 39, 3 (1958).

Homocatechol in an Insect Cuticle

During the course of an investigation into the hardening of the cuticle of the desert locust *Schistocerca gregaria*, 4-methyl-catechol, a substance not previously recorded from insect cuticles, was identified chromatographically in abundant amounts from fully hardened cuticles after acid hydrolysis as well as after alkaline hydrolysis under reducing conditions. This substance, although absent from the soft cuticles newly exposed on moulting, is readily detected after they have been artificially tanned with *B*-(3 : 4-dihydroxyphenyl)acetic acid, indicating decarboxylation of the tanning agent. If *B*-(3 : 4-dihydroxyphenyl)propionic acid is used in place of the acetic derivative to tan the cuticles a compound with similar properties is recovered from the cuticles. This latter compound, which is also present in aqueous extracts of naturally tanned cuticles, has an R_F value, in benzene/acetic acid/water, slightly higher than that of 4-methyl-catechol. It can only be presumed to be 4-ethyl-catechol similarly formed by decarboxylation. *B*-(3 : 4-dihydroxyphenyl)acetic acid and 4-methyl-catechol have also been detected after tanning cuticles with *B*-(3 : 4-dihydroxyphenyl)-propionic acid, and the formation of these compounds seems a clear indication that degradation of *ortho*-diphenolic compounds in an insect cuticle[1] to compounds having shorter side-chains may be accompanied by decarboxylation. Hackman and Todd[2] have already demonstrated the decarboxylation *in vitro* of 3 : 4-dihydroxybenzoic acid in the presence of a plant phenolase.

In the course of this work, it was found that *B*-(3 : 4-dihydroxyphenyl)propionic acid is present in the soft cuticle of *Schistocerca*. This is important, for Hackman[3] has suggested that the degradation of *B*-(3 : 4-dihydroxyphenyl)alanine may not proceed through the propionic derivative.

In the full account of this work, to be published elsewhere, it will be shown that the exocuticle of *Schistocerca*, which differs markedly from the amber exocuticle of other insects[4], is not tanned by *para*-quinones[5,6] but by *ortho*-benzoquinone derived from catechol.

This work was made possible by a grant from Unesco and carried out in the Zoology Department of the University of Manchester. I wish to thank Prof. R. Dennell for the hospitality of his laboratory.

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[1] Richards, A. G., "The Integument of Arthropods" (University of Minnesota Press, 1951).

[2] Hackman, R. H., and Todd, A. R., *Biochem. J.*, 55, 631 (1953).

[3] Hackman, R. H., *Biochem. J.*, 54, 371 (1953).

[4] Malek, S. R. A., *Proc. Roy. Soc.*, B, 149, 557 (1958).

[5] Dennell, R., *Proc. Roy. Soc.*, B, 148, 176 (1958).

[6] Kennaugh, J. H., *J. Insect Physiol.*, 2, 97 (1958).

BACTERIOLOGY

Influence of Cobalt and Zinc Ions on the Growth and Porphyrin Production of *Mycobacterium tuberculosis avium*

It was shown by Baisden[1] that the addition of traces of bacillary ash to a synthetic medium prepared from chemically pure constituents was a very effective stimulant for the growth of *Mycobacteria*, and similar results could be obtained with a mixture of fourteen pure inorganic salts. Various other workers have demonstrated the growth-promoting effect of metal ions, especially zinc[2-4] and manganese[5]. A simpler mixture of pure inorganic salts than that used by Baisden has been described recently by Paterson *et al.*[6]. This is as effective an addition to Dorset's[7] medium for growing large quantities of human type bacilli for the production of tuberculin as is Baisden's.

A substantial stimulation of the growth of *Myco. tuberculosis avium* (strain D_4R) has now been observed, using this trace-element supplement of calcium, cobalt, copper and zinc ions. At the same time, organisms grown on the supplemented medium were usually found to be reddish-brown in colour, and a red pigment which fluoresced strongly in ultra-violet light was readily extracted into cold acetone. Following the procedure used by Todd[8] for the extraction of coproporphyrin III from *Myco. karlinski*, hydrochloric acid solutions of the pigment were obtained having very similar light-absorption characteristics in the range 380–630 mμ to those of coproporphyrin. The porphyrin content of a given mass of organisms was assessed by measuring the optical density at 402·5 mμ of the pigment extracted finally into 10 ml. of 20 per cent hydrochloric acid (v/v concentrated acid). The absorption maximum of the Soret band shifted from 402·5 mμ to 401 mμ on dilution of the extract to 0·15 *N* in hydrochloric acid. This is in agreement with the absorption data of Todd[8] and Jope and O'Brien[9].

In these experiments the organisms were grown on the basic Dorset's medium and on the same medium containing the complete trace element supplement of Paterson *et al.*[6]. Three additional groups of flasks were also prepared in which the supplements simply consisted of either 0·27 p.p.m. cobalt or 1·07 p.p.m. copper or 6·04 p.p.m. zinc. After periods of 33–53 days after inoculation, the organisms were harvested, the yields were determined (expressed as bacillary dry weight) and the porphyrin contents of the bacilli estimated (Table 1).

The results of the experiments show that after 45 days incubation a supplement of calcium, copper, cobalt and zinc stimulated the growth of *Myco. tuberculosis avium* three-fold and at the same time the production of porphyrin (which was maximal after 45 days) was increased about 62 times. The effect of adding the same amount of zinc or cobalt to the medium separately was also to stimulate growth and porphyrin production, but in either case the effect was only about half as great as that induced by the full supplement. Copper alone had no effect either on the yield of organisms obtained or on the amount of porphyrin produced. Thus, the stimulating effect of the trace element supplement of Paterson *et al.*[6] on the growth and free porphyrin production of *Mycobacteria* is largely if not solely due to zinc and cobalt.

Table 1. EFFECT OF COBALT, COPPER AND ZINC IONS ON GROWTH AND PORPHYRIN PRODUCTION OF *Myco. tuberculosis avium* (STRAIN D_4R)

Incubation period. (Days at 37° C.)	Additions* to medium	Yield of organisms (gm.)	Amount of porphyrin extracted. Optical density of 10 ml. hydrogen chloride solution at 402·5 mμ
33	Basic medium	10·0	1·100
38		9·3†	0·915
45		7·9†	0·609
48		8·2†	0·273
33	Full supplement of calcium, cobalt, copper and zinc (ref. 6)	16·0	8·70
41		22·3	11·9
45		23·7	38·2
53		19·6	13·8
42	6·04 p.p.m. zinc	17·1	27·6
47		13·7	21·8
49		13·6	23·3
38	0·27 p.p.m. cobalt	18·0	17·1
42		17·9	16·5
47		17·1	19·9
40	1·07 p.p.m. copper	6·6†	0·441
46		8·6†	0·790
52		7·9†	0·534

* Each flask contained 1 litre of a modified Dorset's synthetic medium with or without trace element additives.

† Pellicle had sunk. All others remained floating during the experiment.

The isolation of the porphyrin pigment and its identification as coproporphyrin III will be described elsewhere.

I wish to acknowledge the permission of the Director of the Central Veterinary Laboratory to publish these preliminary results.

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[1] Baisden, L. A., *Amer. J. Vet. Res.*, 12, 254 (1951).

[2] Henley, R. R., *Amer. J. Vet. Res.*, 1, 25 (1940).

[3] Dekker, T., and Hultema, H., *Nature*, 182, 1387 (1958).

[4] Drea, W. F., *Amer. Rev. Tuberc.*, 74, 145 (1956).

[5] Willison, E. H., Bingenheimer, J., and Rosenthal, S. R., *Ann. Inst. Pasteur*, 94, 49 (1958).

[6] Paterson, A. B., Wright, E. C., and Patterson, D. S. P., *Tubercle* Lond., 39, 275 (1958).

[7] Dorset, M., *J. Amer. Vet. Med. Assoc.*, 84, 439 (1934).

[8] Todd, C. M., *Biochem. J.*, 45, 386 (1949).

[9] Jope, E. M., and O'Brien, J. R. P., *Biochem. J.*, 39, 239 (1945).

Antigens of Spheroplast Membrane Preparations from *Escherichia coli* B

Vennes and Gerhardt[1] demonstrated quite conclusively by serological means that no trace of cell-wall antigens remained attached to the protoplast membranes prepared by lysozyme treatment of *Bacillus megaterium*. Because of the differences in the chemical composition of walls of Gram-positive and Gram-negative bacteria it has been suggested that the osmotically fragile, spherical bodies produced from Gram-negative bacteria by lysozyme treatment or induced by penicillin action have only partially lost their cell-wall material[2]. Salton and Shafa[3] have shown that some cell-wall components remain in spheroplasts prepared by the penicillin method from two Gram-negative species. Their results indicated that the lipo-protein components remained attached to the spheroplasts. The wall preparations from the spheroplasts were also agglutinated by a cell-wall antiserum.

In the present investigation spheroplasts of *Escherichia coli* B were produced by the lysozyme–

versene treatment of Repaske[4]. With the aid of serological techniques it has been shown that (*a*) cell-wall antigens remain in carefully washed membrane preparations from the spheroplasts and (*b*) there are specific antigens of the spheroplast membrane which are not present in detectable amounts in cell-wall preparations.

The bacteria were grown in a glucose-salts medium[5] and harvested by centrifugation in the cold at the end of the logarithmic phase of growth. Cell walls were obtained by differential centrifugation following disintegration of the cells in a Hughes's press[6] at −20° C. The disintegrate was centrifuged for 15 min. at 25,000*g*. Gentle removal of the surface layer yielded wall preparations that could be purified with ease.

Spheroplasts were prepared by lysozyme–versene treatment according to Repaske[4]. The spheroplasts were centrifuged in the cold and the pellet suspended in 0·01 *M* phosphate buffer at *p*H 7 to lyse the spheroplasts. Deoxyribonuclease (0 05 mgm./ml.) was added to facilitate homogenization.

The resulting preparations of walls and spheroplast ghosts were washed twice in *M* sodium chloride and twice in 0·1 *M* phosphate buffer at *p*H 7, with intervening centrifugations at 25,000*g* for 15 min. The homogeneity of the preparations was controlled by phase contrast microscopic examination.

Immune sera against the cell wall and spheroplast membrane preparations were obtained by immunizing two white rabbits for each of the antigen preparations. The animals were inoculated intravenously twice a week over a period of nine weeks. The initial dosage of 2 mgm. was increased threefold during this nine-week period. Following one week of rest from antigen inoculations the animals were killed by bleeding. The 50 per cent end-point method of complement fixation[7] was used in assaying the immunological reactions. The readings were made visually to 10 per cent accuracy.

Cell walls, spheroplast membranes and whole cells were used as antigens in the reactions and each immune serum was tested against all three in the complement fixation test. The two antisera had the same titre against the spheroplast membrane antigen, and the same titre was obtained when antiserum to spheroplast membranes was tested against cell-wall antigen. The reaction between cell walls and homologous antiserum was slightly weaker. The results are indicated in Table 1.

Table 1. TITRES IN THE COMPLEMENT FIXATION TEST OF WHOLE CELLS AND ISOLATED STRUCTURES OF *Escherichia coli* B

Antigen	Antiserum to	
	Spheroplast membranes	Cell walls
Spheroplast membranes	3,840	3,840
Cell walls	3,840	1,920
Whole cells	1,920	1,920

The antiserum to the spheroplast membrane preparation was absorbed with an excess of whole-cell antigen, prepared by heating a cell suspension to 100° C. for 2 hr. The method of absorption followed the conventional procedure. After each absorption the serum was tested against cell wall and spheroplast membrane preparations. The process was repeated until one of the antigens failed to give an inhibition of the hæmolysis.

As can be seen in Table 2, it was possible to remove the antibodies against cell-wall antigens by absorption with whole cells, as indicated by the complement fixation test. However, the antiserum still showed a positive reaction after nine absorptions, when tested against the spheroplast membrane preparation. This means that there are antigenic materials, specific for the spheroplast membranes, which are not present in cell-wall preparations.

Table 2. REACTIONS OF AN ANTISERUM AGAINST A SPHEROPLAST MEMBRANE PREPARATION ABSORBED WITH HEAT-TREATED WHOLE CELLS

The figures in the left-hand column indicate the number of absorptions

Absorption number	Absorbed antiserum tested against	
	Spheroplast membranes	Cell walls
0	3,840	3,840
1	1,920	1,920
2	960	480
4	240	60
6	120	15
8	120	0
9	120	0

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[1] Vennes, J W, and Gerhardt, Ph, *J Bacteriol*, **77**, 581 (1950).
[2] Weibull, C., "Ann. Rev. Microbiol.", **12**, 1 (1958)
[3] Salton, M. R. J., and Shafa, F., *Nature*, **181**, 1321 (1958).
[4] Repaske, R, *Biochim. Biophys. Acta*, **30**, 225 (1958).
[5] Hook, A. E., *et al.*, *J. Biol. Chem.*, **165**, 241 (1946).
[6] Hughes, D E., *Brit. J. Exp. Path*, **32**, 97 (1951).
[7] Kabat, E A., and Mayer, M. M., "Experimental Immunochemistry" (Charles C Thomas, Springfield, Ill., 1948).

Elongation of a Leprosy Bacillus (*Mycobacterium lepraemurium*) in a Cell-free Medium

THE bacillus of human leprosy (*Mycobacterium leprae*) was one of the first bacteria to be identified as the causative organism of a disease; but it remains to this day among the very few that have failed to grow in any type of culture medium. There is indeed doubt as to whether it has ever even been transmitted to an experimental animal. Much of the laboratory work on leprosy has therefore been concerned with the only other closely related organism, the rat leprosy bacillus (*Mycobacterium lepraemurium*), which infects rats and mice, causing disease having many of the characteristics of human leprosy; in common with *M. leprae* it is unusually slow-growing in the body, with at least 10 days elapsing between each generation.

An important advance is the recent observation of limited multiplication of *M. lepraemurium* in tissue culture[1–3], but the organism, like *M. leprae*, has remained uncultivated so far in any cell-free medium, despite a vast, painstaking effort by many workers, using the most varied media[4,5]. Moreover, studies on the respiratory metabolism of *M. lepraemurium* have shown an almost complete lack of response to the many substances tested; even albumin and yeast and liver extracts, which do stimulate respiration, are apparently not themselves oxidized, nor do they promote utilization of other substrates[5].

For some time, workers in these laboratories have found it possible with the electron microscope to distinguish, under certain conditions, a completely

Table 1. MEAN LENGTHS AND PERCENTAGE DEGENERATION OF *Mycobacterium lepraemurium* AFTER INCUBATION AT 37° C. IN VARIOUS MEDIA
Mean length of bacilli after up to 24 hr. incubation (base line) = 1·64 ± 0·01μ, with 5 per cent degenerate

	Medium	Days incubated							
		7		14		28		63	
		Mean length (μ)	Degenerate (per cent)	Mean length (μ)	Degenerate (per cent)	Mean length (μ)	Degenerate (per cent)	Mean length (μ)	Degenerate (per cent)
A	Water	—	—	1·58	(*50*)	1·57	(*50*)	—	—
B	Phosphate buffer	—	—	1·55	(*50*)	1·59	(*80*)	—	—
C	As *B* + 20 per cent sucrose	—	—	—	—	1·64	(*90*)	—	—
D	Nutrient	1·75	(*10*)	1·91*	(*10*)	2·14*	(*80*)	2·24*	(*80*)
E	As *D* + sugar:								
*E*1	+ 10 per cent sucrose	—	—	2·85*	(*10*)	3·12*	(*20*)	3·44*	(*95*)
*E*2	+ 20 per cent sucrose	1·79	(*10*)	2·21*	(*10*)	2·42*	(*30*)	3·12*	(*95*)
*E*3	+ 8 per cent glucose	—	—	2·44*	(*5*)	2·70*	(*10*)	2·88*	(*80*)
F	As *E*1 + isoniazid	—	—	1·70†	(*20*)	1·64†	(*40*)	—	—

The table gives the summated results derived from all experiments performed.

* The difference between these figures and the base line length (1·64μ), as well as between the figures for *D* and all *E*, far exceed any possible chance fluctuation ($P \ll 0·0001$).

† These figures are not significantly above the base line length of 1·64μ ($P > 0·3$); they are lower than the lengths of *E*1 by an amount far exceeding any possible chance fluctuation ($P \ll 0·0001$).

degenerate form of *M. lepraemurium* that is no longer able to produce disease[6]. In this way we have found that in the conventional culture media the bacilli not only fail to multiply but also are degenerate after a few weeks of incubation at 37° C. In one of our experiments, however, involving a liquid nutrient medium of the type used for culturing tubercle bacilli, with 20 per cent sucrose added in the hope of achieving a beneficial stabilizing effect, electron microscopy at 2 months revealed that, although the bacilli had degenerated, a number looked unusually long. It seemed possible that before death some actual, if limited, growth might have occurred in this medium. We therefore followed up this observation by investigating the frequency distributions of the lengths of rat leprosy bacilli after varying times of incubation in different media.

Bacilli of *M. lepraemurium*, partly freed from tissue components, were provided by our colleague Dr. R. J. W. Rees from the homogenized livers of intravenously infected mice. The bacilli were added to the following liquid media, contained in 5-ml. amounts in 25-mm. diameter test-tubes, to make final concentrations of about 5×10^7 bacilli/ml.: (*A*) distilled water; (*B*) phosphate buffer 0·01 *M*, *p*H 7·0; (*C*) phosphate buffer plus sucrose 20 per cent (w/v); (*D*) a basal nutrient medium composed of Difco 'Casamino' acids, 2·5 gm.; asparagine, 0·3 gm.; anhydrous disodium hydrogen phosphate, 2·5 gm.; potassium dihydrogen phosphate, 1·0 gm.; sodium citrate, 1·5 gm.; crystalline magnesium sulphate, 0·6 gm.; glycerol, 25 ml.; distilled water to 1,000 ml. To this basal medium, sterilized by autoclaving, bovine plasma albumin (fraction V), sterilized by filtration, was added to make 0·25 per cent; (*E*) as (*D*), plus sucrose or glucose (sterilized by autoclaving in high concentrations) to give a range from 1 to 20 per cent (w/v), or 0·5–8 per cent (w/v), respectively; (*F*) as (*E*) (with 10 per cent sucrose), plus isoniazid 5 μgm./ml.

The bacillary suspensions in these media were incubated at 37° C. Some were fixed within 24 hr. (base line) by adding formaldehyde to make 2 per cent, while others were incubated for various periods and then similarly fixed. Samples were examined in the electron microscope, set at a magnification of 10,000 by means of standard 0·26-μ diameter spheres of polystyrene latex. A series of concentric circles, the circumferences of which were spaced by 0·5 cm., drawn on the fluorescent screen on which the final image was seen, allowed the lengths of the bacilli to be grouped in intervals of 0·5μ. The lengths of 100 or more bacilli from each sample were measured in this way, and the frequency distributions of lengths and their means were calculated. At the same time the proportion of these bacilli appearing completely degenerate was estimated, giving a rough minimum value for a non-viable count. Of 1,700 bacilli from six samples fixed within 24 hr., the lengths of 74 per cent lay between 1·0 and 2·0μ, and 92 per cent between 1·0 and 2·5μ; the mean was 1·64 ± 0·01μ—a figure very similar to that already reported from the more difficult measurements made with the light microscope[7]. The average proportion degenerate in these base-line samples was 5 per cent.

The mean lengths after incubation (Table 1) indicate no elongation in the three non-nutrient media (*A*, *B* and *C*), compared with the base line at 24 hr.; and degeneration was rapid. On the other hand, a small amount of lengthening could occur even in the ordinary nutrient culture medium (*D*), though again degeneration was fairly rapid. However, in the same medium with 10 per cent sucrose or 8 per cent glucose (*E*), the mean length nearly doubled, and the proportion of bacilli longer than 2·5μ rose from 6 to 67 per cent (sucrose) and to 51 per cent (glucose) in a month (the greater part of the increase being in the first 2 weeks). Moreover, the appearances of degeneration were distinctly delayed, though occurring ultimately and presumably terminating the lengthening process. The contrast in increased length and slowed degeneration between the sugar-containing medium and the same medium alone was evident through the whole range of concentrations of sucrose from 1 to 20 per cent (optimal at 10 per cent), and with 4 and 8 per cent glucose.

The effect of the anti-leprosy drug isoniazid (medium *F*) in preventing the lengthening excludes the possibility that we are witnessing only a passive stretching. Moreover, the electron density (and therefore the weight density) of the bacilli was unchanged in the long bacilli with, if anything, a slight increase in width. This again points to a real increase in bacterial protoplasm in the cultures. We now, therefore, regard the failure to obtain multiplication of the rat leprosy bacillus in culture medium as being due to a failure of the bacilli to divide, and not to their complete inability to metabolize and grow. If means could be found to encourage division, the culture of leprosy bacilli in cell-free media might at last become possible.

We are indebted to Dr. R. J. W. Rees for providing the *M. lepraemurium* and for his interest and advice; and to Miss B. Cooling for undertaking many of the length determinations.

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[1] Rees, R. J. W., and Wong, P. C., *Nature*, 181, 359 (1958).
[2] Garbutt, E. W., Rees, R. J. W., and Barr, Y. M., *Lancet*, 2, 127 (1958).
[3] Wallace, J. H., Elek, S. D., and Hanks, J. H., *Proc. Soc. Exp. Biol.*, 97, 101 (1958).
[4] Eddy, B. E., *Internat. J. Leprosy*, 5, 31 (1937).
[5] Gray, C. T., *J. Bact.*, 64, 305 (1952).
[6] McFadzean, J. A., and Valentine, R. C., *Trans. Roy. Soc. Trop. Med. Hyg.*, 53, 414 (1959).
[7] Hilson, G. R. F., and Elek, S. D., *Internat. J. Leprosy*, 25, 380 (1957).

GENETICS

Conditioned Lines of Flax

IT was reported[1] that when different combinations of nitrogen, phosphorus and potassium fertilizer treatments were applied to plants of a single inbred variety of flax the large differences in plant size produced by the treatments were transmitted unchanged to their offspring and to the second and third generations. When a small type so obtained was crossed reciprocally with a large type the effect was transmitted through the male parent almost to the same extent as through the female. When the two types were reciprocally grafted, both types were identifiable in the stock and scion. It was concluded from this that the environment had induced at least semi-permanent changes in the nucleus or cytoplasm or in both. The following summarizes some of the results from further experiments, carried out in 1958, on the second, third and fourth generations of two extreme types, *NK* and *NPK*. *NK* refers to the descendants of plants treated in 1954 with nitrogen and potassium, and *NPK* to the descendants of plants treated in 1954 with nitrogen, phosphorus and potassium.

In the fourth generation the *NPK* plants were still several times the size of the *NK* plants and there was no evidence over all four generations that the difference between them was diminishing. Seed taken from the stock and scion of the reciprocal grafts of *NPK* and *NK* made in the previous year again produced plants of these two distinct types, and they were identical with plants grown from seed from ungrafted plants, or from plants with scions grafted on to like stocks. In all cases the plants were highly uniform, and the two types would appear therefore to be very stable.

Further reciprocal crossings of the two types show the transmission to be equilinear. It seems that the small deviation towards the female parent previously reported was a maternal effect due to those crosses being made between types growing in their respective environments in which they were conditioned. The F_2 showed more variability than the F_1 though no significant increase was obtained with the somewhat limited number of plants grown. A more extensive study is proceeding.

The F_1's may show dominance depending on the environment in which they are grown, but heterosis occurred only in the F_1 seed weight. These departures from the mid-parent value do not necessarily mean that the primary effect of the environmental conditioning has been the induction of chemical changes on the chromosomes giving rise to genes with dominance relations. Purely quantitative cytoplasmic differences, for example, if transmitted equally through the male and female gametes, could produce an F_1 which differed from the mean of the parents in certain environments, and this may be more marked in some characters due to the interplay of physiological processes. Even so, whether the primary differences have arisen in the cytoplasm or in the nucleus they, together with the external factors, form the genic environment and it is the interaction between them all that must be resolved in a final analysis.

There was a significant difference between the log growth-rates of the two types but not between the log growth-rates of plants having the different fertilizers applied directly to them, although the final weight difference was of the same order in both sets. There are, therefore, fundamental metabolic differences between the two conditioned types, such as might be found between two different genotypes.

The first generation of a diallel cross between *NK* and *NPK* conditioned types and four other varieties, two of which were flax and two linseed, was analysed for a number of characters. They included plant weight, side-shoot weight, centre-shoot weight, height and length of side shoots. The most revealing picture was given by the array covariance/variance graphs using the method of genetic analysis of diallel tables of Jinks[2] and Hayman[3]. With each character, the points for the *NK* and *NPK* types on the graph were close together, although the points representing the other varieties changed their positions with the different characters analysed. It would appear from this that the *NK* and *NPK* types are identical in having similar dominant genes. They were also found to be identical in showing the same non-allelic interaction with one of the linseed varieties in side-shoot weight. Interpreted in another way, suppose the variety which was conditioned were entered twice in the diallel table, then a constant amount added to all the cells of the male and female arrays of one of the entries, to represent the larger (*NPK*) type, would give just such a result, for the addition of a constant amount would make no difference to the array variance or covariance. The similarity between the two types in showing no dominance or interaction differences might be taken as further evidence for the absence of residual genetic variability in the conditioned variety, but complete additiveness in effect of the factors responsible for the difference need not necessarily be expected in all environments for the reasons given above.

Finally, the range of types produced by the different combinations of fertilizer treatments of which the *NK* and *NPK* types are the extremes, resembles the continuous range of variation that might be expected from a genetically determined quantitative character, but the relationship between the two systems involved is as yet unknown.

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[1] Durrant, A., *Nature*, 175, 560 (1958).
[2] Jinks, J. L., *Genetics*, 39, 767 (1954).
[3] Hayman, B. I., *Genetics*, 39, 789 (1954).

FORTHCOMING EVENTS

Monday, January 4

Royal Institute of Chemistry (joint meeting with the Society of Chemical Industry, at 14 Belgrave Square, London, S.W.1), at 6.30 p.m.—Dr. T. H. Blakeley and Dr. A. E. S. White: "The Improvement of Refractories by Incorporation of Metals".

Tuesday, January 5

Institution of Chemical Engineers (at the Geological Society, Burlington House, Piccadilly, London, W.1), at 5.30 p.m.—Mr. J. A. R. Bennett, Mr. J. G. Collier and Mr. H. R. C. Pratt: "Heat Transfer to Two-phase Gas/Liquid Systems—Part 1"; Mr. J. G. Collier, Mr. J. D. Thornton and Mr. J. A. R. Bennett. "Pressure Drop and Hold-up in Two-phase Flow".

Institution of Electrical Engineers, Measurement, Supply and Utilization Sections (at Savoy Place, London, W.C.2), at 5.30 p.m.—Mr. H. J. H. Sketch, Mr. P. A Shaw and Mr. R. J. K. Splatt · "A New Method for Observing the Phenomena of Commutation"; Prof. J. C. Prescott and Mr. A. K. El-Kharashi · "A Method of Measuring Self-Inductances Applicable to Large Electrical Machines".

British Computer Society, London Branch (at the Northampton College of Advanced Technology, St. John Street, London, E.C.1), at 6.15 p m. —Mr. R. L. Sutton: "The First Year's Experience with a Large Computer in a Life Assurance Office".

Society of Chemical Industry, Plastics and Polymer Group (at 14 Belgrave Square, London, S.W.1), at 6.30 p.m.—Dr. E. C. A. Horner. "P.V.C. Degradation and Action of Stabilizers".

Textile Institute (at the Chemical Society, Burlington House, Piccadilly, London, W.1), at 6.30 p.m.—Dr. C. S. Whewell: "Shrinkage of Textile Fabrics".

Royal Photographic Society of Great Britain (at 16 Princes Gate, London, S.W.7), at 7 p.m.—Prof. S. Tolansky, F.R S: "Optical Illusions".

Tuesday, January 5—Thursday, January 7

Institution of Mechanical Engineers (at 1 Birdcage Walk, Westminster, London, S.W.1)—Symposium on "Recent Mechanical Engineering Developments in Automatic Control".

Wednesday, January 6

Institute of Petroleum (at 61 New Cavendish Street, London, W.1), at 2.30 p.m —Dr. C. M. Cawley: "Flow Properties of Admiralty Fuel Oils".

Royal Society of Arts (at John Adam Street, Adelphi, London, W.C 2), at 2.30 p m.—Dr. J. D. Carthy "Animal Senses and Reactions". (Last of two Juvenile Lectures.)

Geological Society of London (at Burlington House, Piccadilly, London, W.1), at 5 p m.—Mrs. D. S Peake "Glacial Changes in the Alyn River System and Their Significance in the Glaciology of the North Welsh Border".

Royal Meteorological Society (at 49 Cromwell Road, London, S.W 7), at 5.30 p.m.—Mr. C. E. Wallington: "Mountain Waves".

British Institution of Radio Engineers (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 6.30 p.m.—Prof. D. G. Tucker "Some New Possibilities in Civil Underwater Echo-ranging—Current Research at the University of Birmingham".

Thursday, January 7

Royal Aeronautical Society (at the Royal Society of Arts, John Adam Street, Adelphi, London, W.C.2), at 3 p.m.—Dr. G. Melvill Jones: "Flying and the Human Machine" (Young People's Lecture).

Institute of Refrigeration (at the Institute of Marine Engineers, The Memorial Building, 76 Mark Lane, London, E.C.3), at 5.30 p.m.—Mr. H. Heckmatt "Some Lubricating Problems of Refrigerating Machines using Refrigerant 12".

Institution of Electrical Engineers (at Savoy Place, London, W.C.2), at 5.30 p.m.—Mr H. G Bell "The Protection of Electrical Systems" (Second Hunter Memorial Lecture).

Society of Instrument Technology (at Manson House, 26 Portland Place, London, W 1), at 7 p m —Mr. E. J. Sumner "The Production and Assimilation of Meteorological Observations".

Friday, January 8

Institution of Electrical Engineers, Medical Electronics Discussion Group (at Savoy Place, London, W.C.2), at 6 p.m.—Discussion on "Electrical and Electronic Techniques in Respiratory Research", opened by Prof. R. Woolmer and Mr. D. W. Hill.

Society of Dyers and Colourists (at the Royal Society, Burlington House, Piccadilly, London, W.1), at 6 p m.—Mr. H. W. Seaman: "New Work on the Dyeing of Nylon with Disperse Dyes".

Antiquarian Horological Society (at the Science Museum, South Kensington, London, S.W.7), at 7 p.m.—Mr. Douglas Hughes "Bells and Bellfounding".

Friday, January 8—Saturday, January 9

Association of Applied Biologists (joint meeting with the British Mycological Society, at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 10.15 a.m. daily—Meeting on "The Nature and Exploitation of Crop Plant Resistance to Disease".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Lecturers/Senior Lecturers (with a first- or second-class honours degree in mathematics or engineering, or at least a second-class general honours degree including mathematics) in Mathematics at the Royal Military Academy, Sandhurst—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. A.568/9A (January 10).

Senior Lecturers or Lecturers (with a good honours degree and relevant teaching experience) in Mathematics and Physics at the Nigerian College of Arts, Science and Technology—The Council for Overseas Colleges, 12 Lincoln's Inn Fields, London, W.C.2 (January 11).

Lecturer/Senior Lecturer in Nutrition, to undertake teaching and research in livestock nutrition in the Department of Nutrition and Chemical Pathology, Faculty of Rural Science, University of New England, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia, January 20).

Babcock and Wilcox Fellow in Nuclear Engineering—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1 (January 30).

Plant Breeder and Geneticist (honours graduate in science or agricultural science (or with equivalent qualifications) with two or more years of postgraduate research experience) at the Tobacco Research Institute, Commonwealth Scientific and Industrial Organization, Mareeba, North Queensland, to take charge of a programme of plant breeding and genetical investigations which has been in progress at Mareeba for three seasons—Chief Scientific Liaison Officer, Australian Scientific Liaison Office, Africa House, Kingsway, London, W.C.2, quoting Appointment No. 815/21 (January 30).

Senior Tutor-Demonstrator in Physics at the University of Sydney, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia, January 30).

Chair of Physiology in the London Hospital Medical College—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 1).

Director (experienced administrator with good scientific attainments, a sound knowledge of arable farming and previous experience in agricultural experimental work)—The Secretary, Norfolk Agricultural Station, Sprowston, Norwich, Norfolk (February 1).

Lecturer (with a good university degree and experienced in the teaching of physics and/or mathematics in schools) in Education—The Secretary and Registrar, The University, Southampton (February 1).

Assistant Trials Officer (preferably with practical experience in crop production), to work at Leeds University Farm, Headley Hall, Yorks—The Secretary, National Institute of Agricultural Botany, Huntingdon Road, Cambridge.

Cell Biologist (with a degree and research experience (preferably a Ph D.) in biology, microbiology or biochemistry), to take part in a research project on the fundamental biology of fibroblasts—Dr J. M Mitchison, Department of Zoology, West Mains Road, Edinburgh 9.

Reader or Consultant in Nuclear Physics—The Principal, Bradford Institute of Technology, Bradford.

Research Assistant (preferably with biochemical, biophysical or physiology training), to join a team engaged in research on the problem of intervertebral disk, herniation and degeneration—The Secretary, Royal Infirmary, Bradford.

Senior Research Assistant (postdoctorate physicist), to undertake research using rockets and satellites; a Senior Research Assistant (post-M.Sc. or Ph.D. mathematician or physicist), to undertake research and to set up and supervise arrangements for data handling (including the use of automatic computers); Junior Assistants (female, with advanced level mathematics, G.C.E.); an Electronic Technician (with initiative, and able to undertake accurate assembly of rocket and satellite electronic systems, and to take part in experiment and testing); and a Draughtsman (with initiative and ability for detail, and willing to take part in practical development work) in the Space Research Group in the Physics Department—The Secretary, University College, Gower Street, London, W.C.1, marking envelope "For the attention of Dr. R L. F. Boyd".

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

British Electrical and Allied Industries Research Association. Technical Report G/XT161: Circuit Interruption in Vacuum By M. P. Reece. Pp. 9+5 figures. 7*s.* 6*d.* net. Technical Report L/T357. The Impulse Breakdown of Uniform-Field Gaps in Air (Interim Report) By R. Galloway. Pp. i+10+21 figures. 12*s.* 6*d* (Leatherhead: British Electrical and Allied Industries Research Association, 1957 and 1958.) [411

Contemporary Physics, Vol. 1. No. 1 (October 1959). Edited by G. R Noakes. (A Journal of Interpretation and Review.) Pp. 80 Subscription price per volume (6 parts). 27*s.* 4 dollars, post free, payable in advance. Price per part, 5*s.*, 75 cents, plus postage (London · Taylor and Francis, Ltd., 1959) [411

Ministry of Education. A Career in Education for University Graduates. Revised edition. Pp. 15. (London: Ministry of Education, 1959) [1011

Building Research Station Digest, No. 126 (September 1959): Mortars for Jointing. Pp. 6. (London: H.M. Stationery Office, 1959.) 4*d*. [1011

The International Association for the Exchange of Students for Technical Experience Twelfth Annual Report, 1959 Pp 72. Report of the International Association for the Exchange of Students for Technical Experience, Great Britain, for year ending September 1959 Pp. 12 (London. International Association for the Exchange of Students for Technical Experience, 1959) [1011

British Rubber Producers' Research Association. Twenty-first Annual Report covering activities for the year 1958. Pp. 50 (London · British Rubber Producers' Research Association, 1959) [1011

Royal Entomological Society of London Handbooks for the Identification of British Insects. Vol. 1, Parts 12 and 13: Mecoptera, Megaloptera and Neuroptera By Lt.-Col F. C. Fraser. Pp. 40 10*s*. Vol 7, Part 2 Hymenoptera Ichneumonoidae. Ichneumonidae, key to subfamilies and Ichneumoninae—1 By J F Perkins Pp 116. 25*s*. (London: Royal Entomological Society, 1959.) [1611

Royal Greenwich Observatory Bulletins No 14: Photoheliographic Results, 1956. Pp 86 (London. H M Stationery Office, 1959) 6*s*. net. [1611

An Audit of Academic Performance a Study of the Prediction of Academic Success in the Queen's University of Belfast. By Michael Forster. Pp 75. (Belfast: The Queen's University, 1959.) 7*s*. 6*d*. [1611

Institution of Chemical Engineers. Education and Training in Chemical Engineering. (Supplement to "Chemical Engineering—a Career".) Pp. 47. (London: Institution of Chemical Engineers, 1959.) [1611

Wye College (University of London). Department of Hop Research Annual Report, 1st April, 1958 to 31st March, 1959 Pp. 90. (Wye, near Ashford Wye College, 1959) [1611

The Operation of Sterilising Autoclaves a Symposium held at Brighton Technical College on May 9, 1959 Pp ii+45. (London · The Pharmaceutical Press, 1959) 7*s*. 6*d*. [1611

The Control of Radioactive Wastes Pp 46. (Cmnd 884.) (London. H.M Stationery Office, 1959) 2*s* 6*d* net [1611

Planning, Vol 25, No. 487 (16 November 1959) The British Shipping Industry Pp 189–220 (London: Political and Economic Planning, 1959.) 4*s* [1611

Annals of the Cape Observatory, Vol. 20: Cape Photographic Catalogue for 1950 0, Zone –56° to –64°. Prepared under the direction of J Jackson and R. H. Stoy. Pp. xxii+150. (London · H.M. Stationery Office, 1958.) 50*s*. net. [1611

Bulletin of the Electronic Forum for Industry, Vol 1, No 1 Pp 16. (London: Electronic Engineering Association, 1959.) [1611

University College of Wales, Aberystwyth Welsh Plant Breeding Station Report, 1st October 1956 to 30th September 1958. Pp 80. (Plas Gogerddan, near Aberystwyth. Welsh Plant Breeding Station, 1959.) 7*s* 6*d*. [1911

Essay Towards Solving a Problem in the Doctrine of Chances. By Thomas Bayes. With a Biographical Note by G A Barnard. (Reproduced from *The Philosophical Transactions* (1763), 53, 370–418, with permission of the Royal Society, in *Biometrika* (1958), Vol. 45, Pp. 293–315) (London: Biometrika Office, University College, 1959.) 5*s*. [1911

British Library of Political and Economic Science (London School of Economics and Political Science, University of London) Annual Report 1958/59. Pp 16 (London British Library of Political and Economic Science, 1959) [1911

Other Countries

Institut Royal des Sciences Naturelles de Belgique Expédition Océanographique Belge dans les Eaux Côtières Africaines de l'Atlantique Sud (1948–1949). Résultats Scientifique. Vol. 3, Fascicule 6: Medusae Mainly from the West Coast of Africa By Paul L. Kramp. Echinides Par Gustave Cherbonnier Pp 59+10 planches (Bruxelles · Insitut Royal des Sciences Naturelles de Belgique, 1959) [411

Companhia de Diamantes de Angola (DIAMANG) Sericos Culturais. Dundo—Lunda—Angola. Publicacoes Culturais No 44: Subsidios para o Conhecimento do Clima da Lunda Resultados das Observacoes Meteorologicas. (Area das Exploracoes da Companhia) Anos de 1952 a 1957. Elaborados por Tomaz Rebelo do Espirito Santo Prefacio do Professor Amorim Ferreira Segundo Volume Apuramentos Mensais e Valores Horarios Estacao Meteorologica do Dundo Pp 685. (Lisboa Companhia de Diamantes de Angola, 1959.) [411

Eastern Region, Nigeria. Annual Report of the Forestry Division, 1958–59 Pp 39 (Enugu: Government Printer, 1959) 1*s* 9*d* [1011

Rensselaer Polytechnic Institute, Troy, New York Catalogue 1959–1960 Undergraduate Courses (Bulletin Vol 58, No 6). Pp 215+12 plates (Troy, New York: Rensselaer Polytechnic Institute, 1959) [1011

Northern Region of Nigeria Annual Report on the Forest Administration of the Northern Region of Nigeria for the year 1957–58 Pp ii+46 (Kaduna: Government Printer; London: Crown Agents for Oversea Governments and Administrations, 1959) 2*s*. net. [1011

Journal of Electroanalytical Chemistry, Vol 1, No 1 (August, 1959) Edited by G. Charlot, J O'M. Bockris and C N. Reilley. Pp. ii+100 Six issues to the volume, approx 500 pages per volume. Subscription price: £6 5*s*. 0*d* or 17.50 dollars, or D.Fl. 66 50 per volume (post free) (Amsterdam: Elsevier Publishing Company; London: D. Van Nostrand Company, Ltd., 1959) [1011

University of California Publications in Geological Sciences. Vol 34, No 3 Tertiary Stratigraphy of the Blairsden Quadrangle, Plumas County, California. By Cordell Durrell Pp 161–192+plates 24–29 1 dollar Vol 34, No. 4 The Lovejoy Formation of Northern California By Cordell Durrell. Pp. 193–220+plate 30. 1 dollar (Berkeley and Los Angeles · University of California Press; London Cambridge University Press, 1959.) [1011

Bechuanaland Protectorate. Geological Survey Department Mineral Resources Report No. 1 The Morupule Coalfield, Palapye Area By O J. van Straten. Pp. vi+50. (Lobatse: Geological Survey, 1959.) 5*s*. [1011

Bulletin of the American Museum of Natural History. Vol. 118, Article 7: Results of the Puritan-American Museum of Natural History Expedition to Western Mexico 7: Corals and Coral Reefs in the Gulf of California. By Donald F. Squires. Pp. 367–432+plates 28–34 (New York · American Museum of Natural History, 1959.) 1 25 dollars [1011

Comité International des Poids et Mesures Procès-Verbaux des Séances 2e Série—Tome 26-A 47e Session—1958 (29 Septembre-30 Octobre), et Comité Consultatif de Thermométrie (5e Session—Juin 1958). Pp. 360+ix. (Paris Gauthier-Villars, 1959) [1011

Cytology, Vol. 1, No. 1. Editor in Chief A S. Trosin. In Russian. Pp 140 (Moscow Akademii Nauk SSSR., 1959) [1011

Union Internationale de Cristallographie Commission des Appareils Cristallographiques. Répertoire de Matérial Cristallographique. (Index of Crystallographic Supplies) 2e Édition. Pp. xxvi+126 (Paris · Société Francaise de Minéralogie et de Cristallographie, 1 rue Victor-Cousin, 1959.) [1011

International Astronomical Union Symposium No. 10 (held in Moscow, 15, 16 August 1958): The Hertzsprung-Russell Diagram Edited by Jesse L. Greenstein. (Reprinted from *Annales d'Astrophysique*, 1959 Supplements, Fascicule No 8) Pp. 127+6 plates. (Paris Centre National de la Recherche Scientifique, 1959). [1011

Western Australia: Geological Survey Bulletin No 109 Miscellaneous Reports for 1954 Pp. 180+17 plates and 23 figures (Perth: Government Printer, 1959.) [1011

Food and Agriculture Organization of the United Nations. FAO Report No 1029 Report to the Government of Indonesia on Horticultural Production and Research. By L B. Thrower. Pp vi+88. (Rome Food and Agriculture Organization of the United Nations, 1959.) [1011

Canada · Department of Mines and Technical Surveys. Geological Survey of Canada Memoir 294. Cornwallis and Little Cornwallis Islands, District of Franklin, Northwest Territories. By R. Thorsteinsson. Pp v+134+8 plates. 1 dollar. Memoir 301 · Fogo Island Map-Area, Newfoundland (2E/9, mainly) By D M. Baird. Pp. vi+42+8 plates. 50 cents (Ottawa: Queen's Printer, 1958 and 1959) [1611

Indian Forest Records (New Series) Wood Seasoning. Vol. 1, No. 2: Studies in Moisture Content, Shrinkage, Swelling and Intersection Point of Mature *Dendrocalamus strictus* (Male Bamboo). By J. Kishen, D. P Ghosh and M A. Rehman Pp. ii+11–30 (Delhi: Manager of Publications, 1959) Rs 1.62, 2*s*. 6*d*. [1611

Smithsonian Institution Proceedings of the United States National Museum Vol 109, No 3412 Marine Mollusca of Point Barrow, Alaska By Nettie MacGinitie Pp 59–208+plates 1–27. Vol 110, No. 3417: A revision of American Bats of the Genera *Euderma* and *Plecotus*. By Charles O Handley, Jr. Pp. 95–246 Vol. 110, No. 3418: A Revision of the Apion Subgenus *Trichapion* Wagner in the New World (Coleoptera: Curculionidae) By David G. Kissinger Pp. 247–389. (Washington, D C. · Smithsonian Institution, 1959.) [1611

Buletinul Institutului Politechnic din Iasi, Serie Noua, Tomul IV (VIII), Fasc 3–4 Pp. xvi+480 (Iasi: Institutul Politechnic, 1958) (1611

Federation of Malaya. Report on Forest Administration for the year 1958 By J. S Smith Pp iv+110+4 plates. (Kuala Lumpur: Government Printer, 1959) 2 50 Malayan dollars [1611

United States Department of Commerce Coast and Geodetic Survey. Technical Bulletin No. 5: Raydist on Georges Bank. By Gilbert R Fish. Pp. ii+25 (Washington, D C. Government Printing Office, 1959.) 25 cents [1611

Food and Agriculture Organization of the United Nations. FAO Agricultural Studies No 43: Efficient Use of Fertilisers Edited by Vladimir Ignatieff and Harold J. Page Pp xxi+855. (Rome · Food and Agriculture Organization of the United Nations; London · H M. Stationery Office, 1958) 20*s*; 4 dollars. [1611

The Industrial Challenge of Nuclear Energy, 3. Stresa Conference, Part 1: Survey of European Programmes, Economics of Nuclear Power, Financing the Programmes. Pp 275 (Paris: Organization for European Economic Co-operation; London: H.M. Stationery Office, 1959) 1,200 francs; 18*s*. 6*d*; 3 dollars; 12 Swiss francs [1611

Darwin Centennial Exhibition of Books—The Private Collection of Robert B Honeyman, Lehigh, Class of 1920 Pp. iii+17. (Bethlehem, Pa. · Lehigh University, 1959.) [1611

National Science Foundation Methodological Aspects of Statistics on Research and Development Costs and Manpower, based on papers presented at a Session of the American Statistical Association Meetings, December 1958 Pp. v+132. (NSF 59–36) (Washington, D.C.: Government Printing Office, 1959.) 1.25 dollars. [1611

United States Department of Agriculture: Agriculture Research Service. Agriculture Information Bulletin No. 205: Salt Tolerance of Vegetable Crops in the West. By Leon Bernstein. Pp. 5. (Washington, D.C.: Government Printing Office, 1959.) 10 cents. [1611

Rubber Research Institute of Malaya. Annual Report 1957. Pp. 95 (Kuala Lumpur · Rubber Research Institute of Malaya, 1959) 3 Malayan dollars. [1611

Kungl Lantbrukshogskolans Annaler, Vol 25. (Annals of the Royal Agricultural College of Sweden) Pp v+527. (Uppsala: Lantbrukshogskolans, 1959.) [1611

Transactions of the American Philosophical Society New Series Vol 49, Part 6: Some Assumptions of Aristotle. By Prof. George Boas Pp. 98 (Philadelphia. American Philosophical Society, 1959.) 2 dollars [1611

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INDUSTRIAL RESEARCH ASSOCIATIONS: SOME TAXATION PROBLEMS

IN discussing recently the report of the Pritchard Committee on the 'rates' charged by local government authorities against charities and kindred bodies in Britain, passing reference was made to the position of the industrial research associations (*Nature*, 184, 1343; 1959). It was stated that the Committee had heard oral evidence from the Parliamentary and Scientific Committee urging that a new statutory relief of 75 per cent should be allowed to industrial research associations. The report notes that under section 449 of the Income Tax Act, 1952, industrial research associations enjoy the same reliefs from income tax as do charities, but the Pritchard Committee submits that the research associations are much more akin to the research establishments of individual firms in industry than to charities. Accordingly, the Committee recommended that the relief considered appropriate for research establishments conducted by individual firms within the curtilage of their industrial hereditaments should be extended to premises of industrial research associations.

The Pritchard Committee, which rightly points out that the treatment for rating of separate hereditaments occupied by individual firms for industrial research is outside its terms of reference, claimed that its view in this matter corresponds to that expressed in evidence by the Department of Scientific and Industrial Research. The report, however, does not indicate the nature of the argument submitted for a new statutory relief of 75 per cent, and in particular, Lieut.-Commander Christopher Powell, secretary of the Parliamentary and Scientific Committee, has pointed out to the Editors that research associations do not at present enjoy the benefit of industrial de-rating. In the view of the Parliamentary and Scientific Committee they should enjoy that benefit, and the figure of 75 per cent recommended was chosen because, when the evidence was submitted, industrial de-rating amounted to 75 per cent.

The memorandum submitted in evidence by the Parliamentary and Scientific Committee points out that research associations do not at present enjoy this benefit of industrial de-rating, because their premises do not fall within the definition of the class of industrial hereditaments comprising 'factories' and 'workshops' in that they are not conducted "by way of trade or for purpose of gain". There is the additional doubt whether 'manual labour' is exercised as required by the definition, when regard is had to the nature of the duties of laboratory workers and research assistants. The research associations might have taken up the matter when legislation affording rating relief to industry was under review, but at the time and until 1948 they were under the impression that they would continue to enjoy the benefits of the Scientific Societies Act.

While it is true that the research associations, which at present enjoy no rating relief of any kind, will obtain 50 per cent relief from rates if the proposals of the Pritchard Report are implemented, so long as that is the percentage of relief accorded to industry, the undoubted anomaly will remain that premises of a commercial concern on which research and development work is carried on may obtain partial exemption from rates, while those of a scientific research association as such obtain none. Associated with this anomaly is the fact that if industrial research and development work by a commercial concern is carried out in a factory hereditament where the predominant activity is production, it can come under the umbrella of industrial de-rating relief afforded to the factory as a whole, but if it is carried out in a separate hereditament full rates have to be borne on that hereditament. The Parliamentary and Scientific Committee submitted that the correct action is to put all premises of the commercial concern in which industrial research and development work proceeds on the same footing regardless of whether or not they form part of the factory.

This is so obviously the logical policy that there can be little doubt that it expresses the general view of the scientist and technologist, and it should be pressed again when the proposals of the Pritchard Committee come before Parliament. Indeed, it is reasonable to claim that research associations have an even stronger claim to rating relief than premises on which a commercial concern conducts research and development work. As the Parliamentary and Scientific Committee urged in its evidence, the research associations do not make profits and cannot pass on the increased cost of their work to the public as can a commercial concern which sells its products. They have educational functions in connexion with training schemes, and their activities educate their member firms in science and technology, sometimes also incidentally, it might be noted, assisting the supply of technically trained staff. Much of the outcome of their work is published in the scientific Press, and their services to technological education should not be overlooked. Notwithstanding that their primary duty is to serve the industries which contribute to their support, their fundamental aim is to see that newly won scientific and technological knowledge is applied throughout British industry.

These public benefits, the extent of which depends on the magnitude of the financial support the associations receive, are every whit as worthy of consideration as their contribution to keeping their industries in the forefront of scientific achievement, which is the fundamental reason why relief from rates is claimed for them. The Parliamentary and Scientific Committee recognizes that privately owned commercial concerns benefit substantially from the work of the research associations, and for that reason does not claim full exemption in the way that it is claimed for scientific societies and institutions. It makes instead the sound claim that the public benefit arising from the work of the research associations justifies relief of 75 per cent from rates, and this relief should stand, independent of whether or not industrial de-rating generally is continued. To that extent, and apart from the specific figure recommended, the Parliamentary and Scientific Committee goes appreciably further than the Pritchard Committee, the report of which does not indicate the full considerations on which the claim is based.

WHITE WILDERNESS

Arctic Wild

By Lois Crisler. Pp. xii+274+60 photographs. (London: Martin Secker and Warburg, Ltd., 1959.) 25*s*. net.

LOIS CRISLER, courageous wife of a professional camera-man engaged in filming arctic wild life for a well-known film company, lived with her husband for two summers and one winter, in tent or flimsy hut, almost entirely alone in the barren wilderness of the Brooks Range in north-western Alaska. During most of this time the two saw no human beings other than the bush pilot who supplied them with the essentials of life at long and irregular intervals, when the weather or the landing conditions did not make his hazardous flights too risky. But they shared a mutual sympathy with, and a dedicated understanding of, the wild life around them. To these two, the tundra, mountains and rivers, in all their changing aspects through the seasons, meant an opportunity to know life much nearer to the primitive than most educated, civilized human beings could ever hope to experience.

To enable them to work unhindered by interference from the occasional Eskimo hunter, and to study and film the undisturbed caribou and wolves, the area in which they lived and worked was made a temporary sanctuary. They themselves carried no firearms. One result of their sojourn in the wilderness, and perhaps the most important one, is this book. It is the work of a writer dedicated to the cause of the conservation of the wild places of the Earth and of the wild animals without the presence of which the face of the Earth remains empty and dead. The picture presented here, in terse but vivid language, is of the wild life of a region that modern man, with his craze for killing and destruction, has still scarcely scratched—the last wilderness of North America. But the frightening hint we are given of what the future holds for this land is depressing enough. As the Crislers flew south from Alaska, their work finished, the mass invasion of the Far North by the military and other personnel engaged in the creation of the *DEW*-Line for the radar defence of the continent was bringing into the wilderness thousands of outsiders to whom any living thing was a target and nothing more, to be killed or wounded indifferently.

To the naturalist, and particularly to the animal behaviourist, this book is a revelation. The main objective of the Crislers was to observe and film, in all possible aspects, the interwoven lives of the caribou in their great migrating herds, and of their attendant predators, the wolves. These are the two most important large mammals of the north. The observations of the writer and her husband form the first clear-cut account of the relationship of the two species. They found that the fit and healthy caribou, even the quite young fawns, were able to outdistance the hunting wolves with comparative ease. In almost all cases investigated it was found that the caribou killed were those hampered by disease, old age, or injury of some kind. The natural role of the wolf, in fact, is as the culler of the unfit, Nature's method of limiting the undue multiplication of the herds. How delicate the balance, and how disastrous the effects of interference with it, is shown by the quoted instance of the effect of wolf 'control' on the Nelchina herd of Alaskan caribou a few years earlier. From an estimated total of 4,000 animals, in ten years the figure, without the limiting factor of wolf predation, had risen to the startling total of about 10,000. The available winter range was no longer sufficient, and the consequent trampling and over-grazing threatened the livelihood of the excessive caribou population. Yielding to wiser counsels, the U.S. Fish and Wildlife Service reversed its former policy and gave complete protection to the wolves in that area. Elsewhere in Alaska a 50-dollar bounty on each wolf sets every man's hand against them and poisons men's minds against their continued existence.

The Crislers' attitude to wolves provides the meat for the most engrossing chapters of this book. They tell of the hand-rearing of wild wolf cubs, and the subsequent sharing of their lives. First two cubs were reared to maturity, and in the second season an entire litter of five, dug out of their den, were brought up by hand and allowed to develop their individual personalities as naturally as possible. In both cases the young wolves regarded the Crislers as their parents, learnt to accompany them on their daily outings on the tundra, to return home after a day's freedom to the pen which was their life's centre, to romp and play, roughly but without malice, with their human substitutes for wolf parents. Naturally this gave unequalled opportunities for detailed study of wolf psychology and behaviour patterns. The results, told with a complete and refreshing absence of the usual animal behaviourist's technical jargon, provide an enthralling story. The excellent photographs, showing adult wolves playing boisterously with their 'foster parents', indulging in communal howling practice with the humans joining in, or accompanying them on their caribou camera hunts, show that the text does not exaggerate the astonishing degree of trust and friendship between wolves and humans. There is rich material here for the student of animal behaviour, and particularly for the student of the origins of the domestication of the dog, to which the wolf is demonstrably so closely akin. The description of the male and female adult wolves, themselves not yet mated, adopting the litter of five young cubs and making it their business to provide all their food in the form of caribou meat carried home in the wolves' stomachs to be disgorged in front of the cubs, is a striking example of the inherent selflessness and gentleness of wolves towards young not even their own.

This is probably the most important study of wolf personality and reactions that has yet been made. I could only wonder at and admire the unafraid trust placed by the Crislers, alone, unarmed, and 90 miles from the nearest help, in these adult wolves, living a practically free and uncontrolled life, yet looking to these two humans for leadership, companionship and play. They admit, casually enough, that at times they had doubts about the reliability of their wolves, but the only moments of trouble and stress came when unfamiliar sights, sounds and persons troubled the animals. To them any break with the familiar pattern of their lives spelt insecurity and danger, and they reacted accordingly.

It will be a very long time before a book as sympathetic to wild life as this (yet without sentimentality or anthropomorphism), and at the same time so readable, is likely to appear. The only criticism I have to make is that the provision of a map would have helped readers to follow the Crislers' journeys and references to Alaskan place-names connected with their work. R. W. HAYMAN

PREHISTORY OF SOUTHERN RHODESIA

Inyanga

Prehistoric Settlements in Southern Rhodesia. By Roger Summers. With contributions by H. B. S. Cooke, P. V. Tobias, H. Wild, J. F. Schofield and K. R. Robinson. Pp. xviii + 336 + 22 plates. (Cambridge: At the University Press, 1958. Published for the Inyanga Research Fund.) 50s. net.

ROGER SUMMERS and his collaborators here describe the fruit of six months field-work under the auspices of the Inyanga Research Fund during 1949–51. Their object was to discover the age and purpose of the stone-walled ruins and terraces that cover between two and three thousand square miles on the Inyanga Highlands of Southern Rhodesia. Summers and his co-workers found that the walled settlements, now in ruin, were built by a prehistoric Iron Age community that displaced the Stone Age inhabitants of Inyanga apparently between the seventh and fourteenth centuries A.D. The reader may ask why we use terms like 'Stone Age' and 'prehistoric' for such recent times, but it must be remembered that, until the nineteenth century, most of Africa south of the Sahara either remained untouched by the great civilizing events that brought writing to the Mediterranean world, or reflected them in part only.

Our previous knowledge of the ruins depended on a few short papers. Now Roger Summers has given us a full monograph, while specialists contribute the latter half of the book on other aspects of the archæology, botany, surface geology, animal and human palæontology of the area. Their enthusiasm carried them beyond the limits of the Iron Age, down into Stone Age depths, for K. R. Robinson contributes a chapter on people who lived in Inyanga long before the Iron Age stone-wall builders arrived there.

Summers's account of the Iron Age at Inyanga is based on no less than thirty-six excavations within an area of 800 square miles. I congratulate him on achieving the aims of the sponsoring Fund, and presenting such an interesting account of the framework of the thriving population that based its way of life on the terraces, trackways, water furrows, stone buildings and pottery it left in this region. Here, too, is an account of the metal-producing techniques that supplied the spears, axes, hoes and knives necessary to the economy and its defence; and also the east coast trade that for centuries poured millions of glass beads and other objects into African Iron Age communities. These beads give some indication of absolute dating, as J. F. Schofield's valuable chapter suggests; but in general these things have an age-range too wide for any but the broadest dating.

P. V. Tobias interprets the two Iron Age skeletal remains as representing a Negro and a Boskopoid–Negro hybrid, anatomical characters found in the present-day Bantu-speaking population in Rhodesia. There may be little doubt that the Iron Age remains in Inyanga are due to their ancestors, and not to any exotic people.

Summers distinguishes two main Iron Age cultures in Inyanga. The earlier cultures may have flourished between the seventh and eighth centuries A.D., but could be as late as the fourteenth century A.D. The scarcity of foreign imports supports the earlier date; indeed, accurate radiocarbon age estimation may show it to be even earlier. Recent radiocarbon dates for Iron Age settlements in Northern Rhodesia suggest that metal-producing farmers may have arrived in the area centuries earlier than our previous conjectural dating based mainly on imported beads.

The later cultures produced the astonishing terraces, stone-lined pits, roadways and water-channels which are such striking features of the Inyanga landscape, as aerial photographs in the book show. These Iron Age people had a thorough grasp of soil conservation, for they made miles of contoured terraces for grain fields and built massive stone walls across gullies to stop soil erosion.

Summers gives a study of the environmental background to Iron Age life, but the reconstruction of climatic change on the basis of erosion and deposition is considered uncertain in view of recent South African research which, in my opinion, has done much to disprove the traditional hypotheses of this kind. Few specialists would accept a chronology based on the hypothetical climatic changes Summers proposes in his concluding chapters.

The archæological remains described in this book are as impressive as Zimbabwe, but it is unfortunate that Summers omits clear comparison with that well-known centre and its associated settlements. I hope he will give us a third book relating Inyanga and Zimbabwe to each other and to all other Iron Age remains in Rhodesia that interest both the archæologist and the general observer of African achievement in the past.

The book reaches the usual high standards of the Cambridge University Press; the line drawings are excellent, but the photographs could have been improved by 'bleeding' the blocks for reproduction.

REVIL MASON

EVOLUTION FOR THE GENERAL READER

Nature and Man's Fate

By Prof. Garrett Hardin. Pp. xii + 375. (New York: Rinehart and Company, Inc., 1959.) 6 dollars.

IN their introductory remarks the publishers of Prof. Hardin's book on evolutionary theory and the course of human evolution describe it as "challenging", "provocative" and "controversial". Even though words of this kind have been used so much that they have lost a great deal of their original force, there is little doubt that "Nature and Man's Fate" will bring pleasure to all those who prefer to see the truth set down in terms of black and white.

The plan of the book is on orthodox lines. The emergence of the theory of evolution as set out by Charles Darwin and A. R. Wallace is preceded by accounts of earlier theories and the intellectual and emotional atmosphere which existed when "The Origin of Species" was published. This is followed by the story of how evolutionary theory gathered momentum and support with the re-discovery of the Mendelian laws of heredity. The latter part of the book explores the significance of genetics in human life and development. In general, the historical and factual part of Hardin's book is remarkably good, mainly because of the interesting and often novel anecdotes and reminiscences which the author produces to support the theories and philosophies of the leading *personalia*.

When Hardin begins to speculate, however, he adopts the overbearing attitude so typical of those who see the world devoid of any shade of grey. Darwin is chided for his lack of mathematics, his support of pangenesis, and his inability "to discover genetics". All Lamarkists are banished from court. "Samuel Butler and St. George Mivart were intensely proud and personally ambitious far beyond their small abilities". It would be interesting to hear Hardin's assessment of a well-known English professor of zoology who advocates that more may be heard of the inheritance of acquired characteristics when the possibilities of cytoplasmic inheritance have been further explored. In his attack on the Soviet attitude to genetics—which is weakened when Hardin adopts the approach of a journalist rather than a man of science—Needham, Bernal and Haldane are accused of something almost verging on intellectual dishonesty. All this to support Hardin's strongly held view that what evolutionary theory and genetic laws have taught us is the essential need for competition if human life is to go on.

It is a pity that the author has spoilt what might have been a good book with over-protestation. "Nature and Man's Fate" should bring him much publicity but little credit. T. H. Hawkins

HEAT TRANSFER AND HEAT EXCHANGERS

Heat Transfer

Vol. 2. By the late Max Jakob. With the technical and editorial assistance of Prof. S. P. Kezios. Pp. xxxii+652. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1957.) 120s. net.

An Introduction to Fluid Mechanics and Heat Transfer

With Applications in Chemical and Mechanical Process Engineering. By Prof. J. M. Kay. Pp. xvi + 309. (Cambridge: At the University Press, 1957.) 37s. 6d.

Heat Exchangers

Applications to Gas Turbines. By Dr. W. Hryniszak. Pp. xii+343. (London: Butterworths Scientific Publications; New York: Academic Press, Inc., 1958.) 63s.; 10 dollars.

THE second volume of the late Prof. M. Jakob's treatise on "Heat Transfer", completed under the guidance of his colleague, Prof. S. P. Kezios, maintains the very high standard set by the first. The first volume left over for consideration the solution of the equations for radiant heat transfer in non-absorbing and absorbing media, a topic which occupies the first quarter of this second volume. The greater part of the book deals with selected fields of application in heat-transfer engineering, discussing first the well-established topics by giving a mathematical treatment of recuperative and regenerative heat exchangers and cooling tower theory. There follows consideration of the newer branches of study in the science of heat transfer, including transpiration cooling and cooling by falling liquid films, heat transfer problems encountered at high fluid velocity through laminar and turbulent boundary layers, and concluding with chapters on heat transfer in liquid metals and in packed columns. A supplement brings up to date some of the material in Volume 1, though none of the many literature references given throughout the book is carried beyond 1953, and indeed those relating to regenerator theory do not go beyond 1940. Presentation of further material is held over to a later edition of the treatise. The two volumes together form an indispensable work of reference for the libraries of all heat transfer laboratories.

Prof. Kay's book is based directly on a university course of lectures in chemical engineering. Picking up this book after Prof. Jackob's two volumes, one wonders how the author has compressed into a modest volume an introduction both to heat transfer and fluid mechanics. This has been achieved by an admirably concise mathematical treatment of his material, but also by omitting entirely consideration of radiant heat transfer and any solution of the unsteady state conduction equations The particular strength of the book lies in the successful manner in which it inter-relates and interweaves the phenomena of fluid flow, mass transfer and heat transfer. The author makes use of vector notation to an extent which need not cause undue difficulty to one to whom it is unfamiliar, though it seems inappropriate to introduce the simple concept of thermal resistance via equations written in vector notation. While there are a few short chapters at the end of the book dealing very briefly with applications in process engineering, the absence of any worthwhile mention of two-phase flow problems is surprising in a book written for chemical engineers. One suspects that this is because their study lends itself less readily to the neat and tidy mathematical treatment which features so largely in this book.

The book by Dr. Hryniszak is in a different category from the two others. While Prof. Jakob's is likely to appeal most to the postgraduate student and research worker and Prof. Kay's to the more able undergraduate, Dr. Hryniszak's would make little appeal to either, for it is essentially a book for the specialist in gas-turbine heat exchangers. Although the non-specialist student might find some interest in the sections dealing with the mechanical aspects of recuperative and regenerative heat exchangers, he would find little attraction in the manner of presentation of the theoretical matter; for in an attempt to keep the mathematics as simple as possible, the theoretical relations are frequently presented as complicated formulæ with little in the way of derivation. Consequently, the contents of the book too often resemble extracts from a design manual, and such manuals seldom make exciting reading. Only the specialist will have the patience and inclination to dig deeper if he wants to compare the author's methods with his own. The volume deals not only with the heat exchanger but also, in optimization procedures, treats it as an integral part of the gas-turbine plant. R. W. Haywood

Pictorial History of Philosophy

By Dagobert D. Runes. Pp. x+406. (New York: Philosophical Library, 1959.) 15 dollars.

THIS handsome volume includes approximately 1,000 portraits, photographs, facsimiles, archæological illustrations and other pictorial material connected in one way or another with philosophy. The net is cast very widely; Goethe, Hitler, Buddha, massacres of the Jews by the Crusaders, photographs of practices by adherents of Yoga, compete for attention with the more conventional practitioners of the craft, such as Aristotle, Cicero, Duns Scotus, Husserl, Wittgenstein and Dewey. Although the principles of exclusion are not obvious, the effect is to have here a collection of philosophical portraits of a range which it would be hard to find elsewhere. Title-pages from important classics are liberally spread throughout the volume, as well as dramatic scenes from the general history of thought and religion, for example, Calvin arguing with Farel, alchemical laboratories, the dying Seneca, Pericles addressing the Athenians. There is less to be said for the literary entries that accompany the illustrations. They vary from a sixth to a third of one page, Kant getting rather less than, say, Carlisle, Croce or Russell, and too frequently mention what is trivial and biographical at the expense of an accurate appraisal of a person's thought. The page facing Russell's portrait shows Stalin, and that with Heidegger has Hitler's portrait: the author makes no bones where his sympathies lie; deeply pro-Jewish, with a full page for "Lincoln, Sage and Seer", and a penchant for the wisdom of the East. Perhaps, though this may introduce an amateurish note, the feelings that emerge are none the worse for it; and if one would not want to learn one's philosophy from it, one must be grateful for the very many rare photographs such as a daguerreotype of Schopenhauer or others of Lloyd Morgan, Alexander, Bradley, Bosanquet; all reproduced with magnificent quality.

GERD BUCHDAHL

Theory and Applications of Nuclear Induction

By Prof. Ajit Kumar Saha and Tara Prasad Das. Pp. x+516. (Calcutta: Saha Institute of Nuclear Physics, 1957.) Rs. 25; 40s.; 6 dollars.

BY and large, there is only one thing wrong with this book—its date of publication. The reviewer's natural sympathy is with Prof. Saha, who completed the task interrupted by his father's death in 1956. But a manuscript completed in 1953 and only slightly revised before receipt in 1959 (although dated 1957). is not in keeping with the pace of physics to-day. The book is written primarily with the experimental scientist in mind; it covers relaxation time and moment measurements, quadrupole effects and chemical shifts in detail. Some mathematical ability and a knowledge of the language of quantum mechanics is assumed, but the arguments are presented in such detail that it should be easy for those learning the subject to follow them. In the authors' own words, "the attitude has been to discuss elaborately the phenomenological theory and the experimental techniques". They have achieved this aim, but it is of course the weakness of phenomenological theory that it covers only the cases discussed; extension and generalization are hazardous. The notable example of this difficulty is in the absence of a treatment of negative-spin temperatures.

The book is well printed, well indexed, and an errata sheet bound into the volume has corrected a good many minor slips. The bibliography is excellent up to 1956, and the book will save a good deal of searching of the literature before that date.

W. M. LOMER

The World is Round

The Story of Man and Maps. By Prof. Frank Debenham. Pp. 100. (London: Macdonald and Co. (Publishers), Ltd., in association with Rathbone Books, 1959.) 50s. net.

"THE World is Round," by Prof. F. Debenham, is "a far cry from those primitive attempts to the drawing of actual maps". The evolution of man's cartographical skill has been achieved by the use of vivid colour reproductions of imaginary world maps and ancient charts. Having traced man's global knowledge to the present day, the author uses specially designed colour relief maps to show the world as we know it to-day. Many unfamiliar sectors of the Earth are revealed and each continent is shown in relation to its neighbour, thus bringing out many points of comparison hitherto unrealized. Prof. Debenham avoids stress on one country more than another, and points out a sociological moral through a geographical fact; the Earth is indeed round, but its centre is not the place where we live.

The pages dealing with the historical background of each country are, in my opinion, out of place in a book of this kind, and tend to detract from its main purpose. From a concise section dealing with map-making instruments through the ages the reader is led to a review of man's latest inventions in mapping under the sea, under the land, and in outer space. This is a beautifully presented book that would not only provide a most useful aid in the school library but also would grace the shelf of any home.

M. CARADINE

La Vie des Colibris

Les Trochilidés. Par Ariane Martin et Anne Musy. (Les Beautés de la Nature—Série Exotique.) Pp. 246+32 planches. (Neuchatel: Éditions Delachaux et Niestlé, 1959.) n.p.

HUMMINGBIRDS, like butterflies and birds' eggs, are natural objects which interest the collector as well as the scientist; they are, as described in this book, "living precious stones" showing a fascinating variety of colour and form within the limits of their distinctive and homogeneous group. Although the object of the series to which this volume belongs is to extol the beauties of Nature, it is the biology rather than the art of hummingbirds which is the theme.

The text consists of a short historical note and chapters on geographical distribution, structure and morphology, temperature regulation, locomotion, food, reproduction and behaviour. These subjects are, of necessity, very briefly summarized. It is not widely known that hummingbirds have a temperature-control mechanism, so that in certain circumstances they can become torpid. Hibernation, in fact, was described in hummingbirds a long time ago.

To those who know little or nothing of these fascinating creatures this book conveys a fair summary of the facts of their biology. The text provides informative reading for the interested layman who, if he understands art, might also appreciate the colour plates.

J. D. MACDONALD

GEOMETRY OF THE STRUCTURE OF MONATOMIC LIQUIDS

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SINCE the work referred to in my communication to *Nature* early in 1959 [1], it has been possible to approach the problem in a more quantitative way by the use of computers and geometrical models. Following up the concept of a liquid as an essentially irregular assembly, I have been able to realize a mathematical model of such an assembly, thanks to the help of Dr. A. D. and Mrs. Booth at Birkbeck College, and of the University of London Computer Unit through the work of my son, Dr. M. J. M. Bernal. A method was devised for computing the co-ordinates of centres of random assemblies of hard spheres with the one condition that no two centres should approach at less than a minimum distance. It proved possible to arrange these in systems with varying densities, the chief interest being, however, in the arrangements of highest density. Later it was possible to build up such arrangements with as many as seventy-five spheres in various ways, to compute from these their mutual energies on the simple Lennard-Jones $-A/r^6 + B/r^{12}$ potential rule and also to determine their effective volumes.

The general picture which emerged was that each particular random arrangement could be altered in absolute scale so as to get a minimum energy for each mutual relative metric position of the points. It could further be modified without preserving these metrical relations but yet maintaining the same topological neighbourhood relations so as to get even lower energy values.

From these minimum energy arrangements it is possible to calculate mean radial distribution functions (Fig. 1). These resembled roughly those obtained experimentally by X-ray and neutron diffraction from monatomic liquids. Dr. Furukawa has shown[2] that nearly all metals show the same distribution function if these are calculated from their observed diffraction curves in the same way. The distribution functions derived from the random arrangement described are similar to this with the significant difference that the first or principal maximum is considerably broader in the model. Now this is exactly what should be expected from the way the model was derived by emphasizing minimum rather than most probable distances of neighbours, for it proved impracticable at this stage to devise a programme which ensured arrangements with a maximum of molecular contact.

To overcome this difficulty, I used a semi-empirical device by building an actual physical model corresponding to the machine-calculated distances. Then, after joining neighbouring points by links of variable length, devised by my technical assistant, Mr. J. Mason, I was able, by a process of manipulation, to reduce most, but by no means all, of the distances between neighbouring points to approximately identical minimal values. This gave a new kind of distribution function (Fig. 1) with much sharper peaks, sharper actually than those given by metallic or molecular liquids (Fig. 1) but more closely approximating to them than in the first model. It also gave a density of packing of 0·61, approaching the value of 0·63 determined by G. D. Scott, (private communication) for irregularly close-packed spheres.

It is evident that this model, by an intermediate degree of equalization of the shorter bonds, could be made to provide radial distribution functions approximating, within experimental limits, to those calculated from diffraction curves. This agreement provides strong support for the principles of construction of the geometrical model proposed for liquid structure. Although, on account of the inherent phase ambiguity of diffraction studies, it is impossible to *prove* the correctness of the model by these means, it equally cannot be disproved, and it must correspond more closely to the real distribution than any model hitherto proposed. Its closest affinity is, in principle, to that of Furukawa[2], who was the first to explain the location of the second peak of the distribution function.

A contemplation of the bond-equalized model led me to the radically new conception of the ***ideal structure of a liquid***, ideal in the sense that in such a structure all the distances between closest neighbours are equal. This ideal corresponds to the ideal structures in crystallography by which, for example, Pauling worked out the structures of the silicates. Actual structures, however, differ, and it is certain that no actual liquid structure can be an ideal one, though it may be close to it.

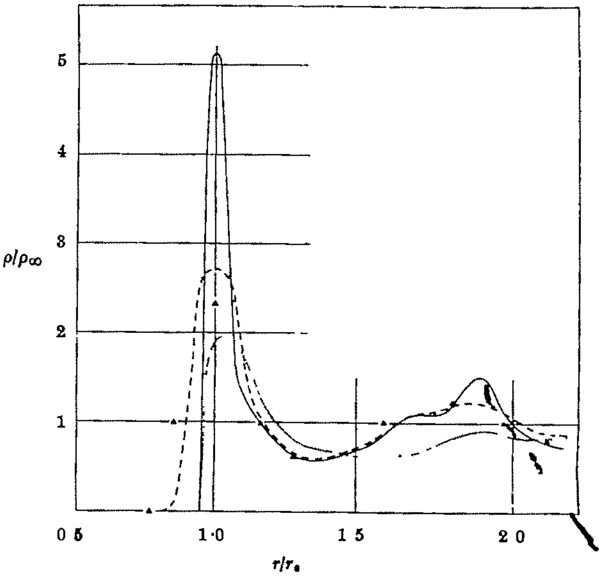

Fig. 1. Density distribution function: ρ/ρ_∞ variation of mean particle density as a function of r/r_0

. . , Derived from calculated random model; ——, derived from squeezed random model; - - - -, calculated for lead by Furukawa (ref. 2); ▲▲▲▲, calculated for liquid argon by neutron diffraction (Henshaw, D. G., *Phys. Rev.*, 105, 976; 1957)

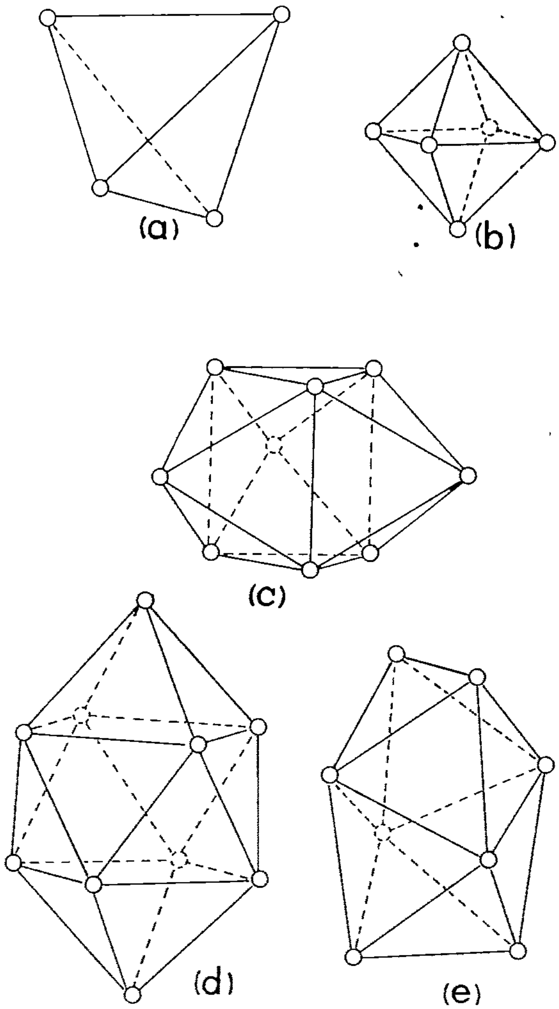

Fig. 2. Forms of holes in ideal equidistant molecule model of a liquid. *a*, Tetrahedron; *b*, octahedron; *c*, trigonal prism capped with three half octahedra; *d*, archimedean antiprism capped with two half octahedra; *e*, tetragonal dodecahedron. (Note prevalence of five-fold surface co-ordination in *c*, *d* and *e*)

Examination of the model showed that it could be considered most profitably as broken down into a set of *polyhedral* holes between molecules. These polyhedra are necessarily small because, by definition, in any coherent *close-packed* liquid there are no holes large enough for another molecule to be inserted. All have in common faces composed of equilateral triangles, or, in other words, *all their edges are equal.* They can, in fact, be derived from five ideal basic shapes: two platonic, two regular archimedean polyhedra, and one of lower symmetry. These are the tetrahedron, the octahedron, the trigonal prism, the archimedean antiprism, and another eight-cornered form, the tetragonal dodecahedron (Fig. 2). These forms can be combined in an unlimited number of ways through the sharing of triangular or square faces (here the half octahedron can appear as a pyramid on a square base). There are no other forms with equal edges, though a relaxation of this rule allows a few other kinds of holes to occur. However, *in general,* such packing cannot fill space without breaking, though to a degree limited to some 15 per cent, the condition of the equality of nearest neighbour distances. The introduction of archimedean polyhedra prevents any long-range order and also prevents a realization of the ideal structure precisely; for the sums of the dihedral angles of the polyhedra can never (except in the case mentioned below) be made exactly to amount to four right angles. In one particular case, that where only tetrahedra and octahedra occur, the packing can be regular, giving rise to varieties of cubic or hexagonal close-packed crystals.

It is possible, therefore, to formulate the geometrical hypothesis of the structure of liquids as follows:

In an *ideal* liquid structure, the molecular centres lie at the apices of a set of empty polyhedra (holes) with equal edges (r_0), or a combination of them. These polyhedra are: platonic regular (1) tetrahedra; (2) octahedra; archimedean (3) trigonal prisms; (4) archimedean antiprisms; and (5) tetragonal dodecahedra. The presence of the three latter forms prevents any long-range order and thus permits fluidity appropriate to temperature, while their proportion determines the configurational volume of the structure at different temperatures. For any temperature the structure is characterized only by the mean value $\overline{N}$ of molecular neighbours, varying from 11 to 3 as the temperature rises, and by the variance (λ) of the intermolecular distance (r_0), which increases with temperature.

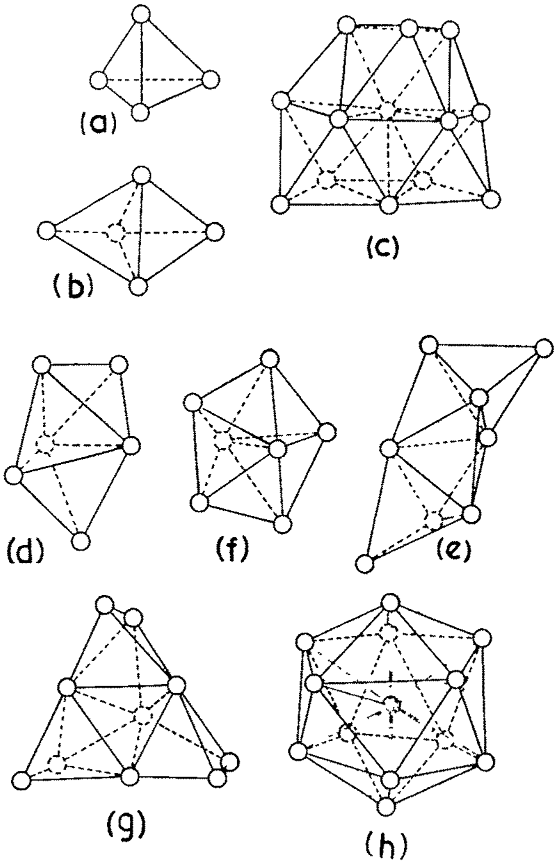

Fig. 3. Crystal nuclei and pseudo-nuclei built from equidistant or quasi-equidistant molecules. Crystal nuclei. *a*, single; *b*, double tetrahedra; *c*, close-packed layers, four tetrahedra and three octahedra. Pseudo nuclei: *d*, three; *e*, four tetrahedral elements of super close-packed spiral chain; *f*, five tetrahedra in pentagonal bipyramid; *g*, seven tetrahedra with elements of three spiral chains; *h*, twenty tetrahedra as fitted in an icosahedron. (In *f* and *h* all the intermolecular distances cannot be precisely equal)

This concept of the ideal structure also opens the way to a real statistic of liquid structure, the relative numbers of the platonic and archimedean polyhedra, depending on the temperature, giving the possibility of determining a mixing entropy to account for the large entropy of the liquid state. This would seem to provide a geometrical basis for Eyring's[3] partition function in his new thermodynamic theory of liquid structure. It can be seen that this geometric view contains the essential features of many earlier theories of liquids, including that of Born and Green[4] based on gas laws, the hole theory of Kirkwood[5] and Frenkel[6] and the crystallite theory of Eyring and others, and points to their eventual reconciliation.

With regard to the last of these, it is apparent from the model that even the most irregular structure contains regions composed of a number of tetrahedra sharing faces. Even though their density may be greater than close packing, as Boerdijk[7] has shown, they are not in general crystalline, but are asymmetric or have forbidden symmetries, such as pentagonal (Fig. 3). They cannot lead to three-dimensional repeat structures and consequently cannot function during the time of their stability as nuclei of crystallization. This may indeed provide the explanation of the phenomena of nucleation and crystal growth in super-cooled liquids. The character of these pseudo-nuclei near their melting point may well be dependent on whether they are formed from the melting of a crystal or from the cooling of a liquid heated to a high temperature. This may also explain some observed hysteresis phenomena in liquids near the melting point. It also implies that with poly-atomic molecules there may be several degrees of melting or break-up of such non-crystallographic aggregates which may help to account for viscosity anomalies with temperature which will appear not as discontinuities but changes of higher order. Further, the existence of such condensations indicates the necessity of fluctuations in density in normal liquids of the dimensions of several interatomic distances as postulated by Furth[8].

A fuller account of this work will be published elsewhere.

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[1] Bernal, J. D., *Nature*, 183, 141 (1959).

[2] Furukawa, K., *Nature*, 184, 1200 (1959); and private communication.

[3] Eyring, H., Ree, T., and Hirai, N., *Proc. U.S. Nat. Acad. Sci.*, 44, 683 (1958) and Fuller, E. J., Ree, T., and Eyring, H., *ibid.*, 45, 1594 (1959).

[4] Born, M., and Green, S., *Proc. Roy. Soc.*, A, 188, 10 (1946).

[5] Kirkwood, J. G., *J. Chem. Phys.*, 3, 300 (1935).

[6] Frenkel, J., "Kinetic Theory of Liquids" (Oxford, 1946).

[7] Boerdijk, A. H., *Philips Res. Rep.*, 7, 303 (1952).

[8] Furth, R., Rheology Abstracts · Account of the Acoustics Group of the Physical Society Meeting at Imperial College, London, September 14–15, 1959 (in the press).

SALTS AND RESPIRATION

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ANIONS and cations of inorganic salts are non-metabolically absorbed by the protoplasm, a process obeying common principles of ion exchange and Donnan equilibrium[1], but metabolic energy is always needed for accumulation of these salts in the vacuoles[2,3]. Even if the initial salt absorption of the protoplasm is a spontaneous process, it is important to realize that the power of salt absorption of the protoplasm, namely, the amount of ion carriers (symbolized as R^- and R^+), is also metabolically regulated[4]. Little is known about the chemical nature of the carriers. They behave as large organic ions and reflect the ionic activity of the protoplasm itself. Normal aerobic respiration is needed for maintenance of the structural qualities of the protoplasm. The activity of the ion carriers is concommittantly more or less abolished if the cells are poisoned by cyanide[4].

From a biochemical point of view the pronounced sensitivity to cyanide points to co-operation of the cytochrome system in the absorption and accumulation of salts. This conclusion had been drawn in 1935[2a]. Later on, direct observations of the response of the respiratory enzymes to presence or absence of salts were made[5]. Rapid measurements of the height of absorption bands of reduced cytochromes a_3, a, c and b of a bundle of wheat roots showed that the bands are high if the roots are 'desalted', that is, thoroughly washed in distilled water, and decrease in height upon addition of 0·001–0·005 M solutions of neutral salts or dissociated salts of organic acids (see Fig. 2). Recent results show a similar response of diphosphopyridine nucleotide and flavoprotein, which are also participating in the respiratory chain. The response to salts is thus confined to the *total* respiratory system, not only to single enzymes.

Discussions of the mechanism of salt accumulation are frequently hampered by ignorance of this distinction between 'initial absorption' and 'continued accumulation'[4,6]. A large number of cells, among them most animal cells and meristematic cells of plants, have much less 'free water' than fully grown cells of plants, in which the sap space may occupy more than 95 per cent of the total volume. The process of continued accumulation is consequently much more dominating in the latter. A question of considerable theoretical interest was raised by recent observations of a sudden stimulation of aerobic respiration upon addition of salts, namely, stimulation occurring in the initial period before the full start of continued accumulation[4]. Because in plant tissues the two phases of absorption are fairly close together in time (the phase of accumulation starts about 15 min. later than the initial absorption), a closer study of the problem required an organism in which salt accumulation takes a subordinate role. Baker's yeast is such an organism. Most inorganic salts pass into the yeast cell, but no significant accumulation above the level in the medium was observed. Yeast is provided with a complete cytochrome system which, in co-operation with pyridine nucleotide and flavoprotein, maintains a fairly intensive aerobic respiration.

The Salt Effect on Baker's Yeast

Suspensions (5–15 per cent) of washed baker's yeast were investigated in the Barcroft–Warburg

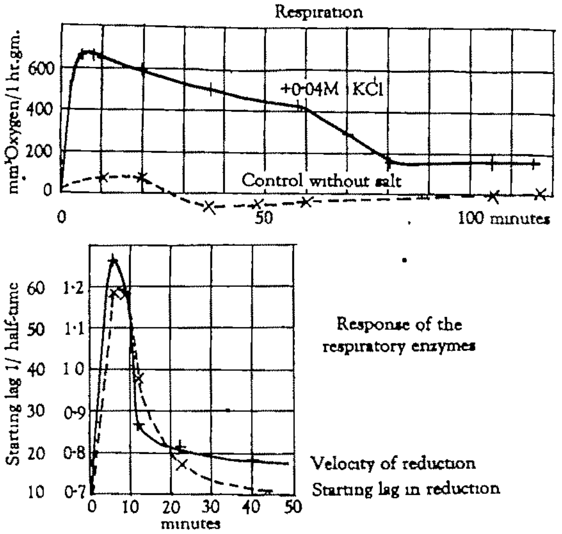

Fig. 1. Response of oxygen consumption (above) and activity of the respiratory enzymes to an addition of 0 04 *M* potassium chloride to a suspension of washed baker's yeast. Identical vessels were investigated, one set without and one with salt. The initial rapid burst of respiration is recognized both in oxygen consumption and reaction of the enzymes

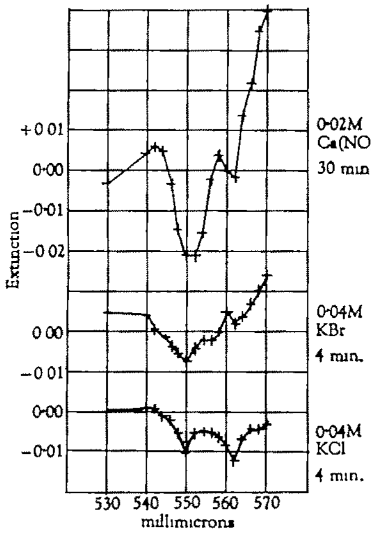

Fig. 2. Response of the cytochromes *c* (550 mμ) and *b* (562 mμ) to an addition of salts Difference spectra were recorded, one quartz vessel (see ref. 7) containing yeast without salt, the other with salt. 1 per cent glucose was added to both vessels in order to obtain 90 per cent reduction from the start. The response on salts implies lowering of the reduced peaks

respirometer and simultaneously in recording spectrophotometers of my own construction[7]. The spectrophotometric observations were carried out according to the procedure developed in my laboratory for studies of the kinetics of the respiratory enzymes, namely, difference spectra were recorded between control (in water) and sample (with salt). Records were made during consecutive time-intervals. For the determination of start lags and apparent reaction velocities of reduction and oxidation, intermittent gassing with nitrogen or air respectively was used, according to a previously elaborated technique[8]. The spectra were in this case recorded by means of a rapid-scanning spectrophotometer.

The response of the oxygen consumption to addition of salts is shown in Table 1 and Fig. 1. Figs. 2, 3 and 4 show the corresponding response of the enzymes.

Table 1. INCREASE OF RESPIRATION ON ADDITION OF SALTS. 2 C.C. 15 PER CENT YEAST SUSPENSION IN EACH WARBURG VESSEL. SALTS ADDED UP TO 0·05–0·20 *N*. TIME OF EXPERIMENT, 110 MIN. RELATIVE VALUES, CONTROL WITHOUT SALT = 100

(*A*) *Series with start in distilled water*						
I. Addition of potassium salts						
Anion:	Cl	Br	I	NO_3	HPO_4	SO_4
	121	122	117	117	127	117
II. Addition of nitrate						
Cation.		K		Ca		Mg
		117		141		140
III. Addition of chloride						
Cation.		K		Ca		NH_4
		121		115		116
(*B*) *Series in 1 per cent glucose*						
I. Addition of potassium salt						
Anion:	Cl	Br	I	NO_3	HPO_4	SO_4
	126	131	133	128	120	124
II. Addition of nitrate						
Cation.		K		Ca		Mg
		127		120		128
III. Addition of chloride						
Cation.		K		Ca		Na
		126		144		141
IV. Addition of sulphate						
Cation.		K		Mg		
		126		123		

Table 1 shows that salts increase the respiration 20–30 per cent both in distilled water and in 1 per cent glucose, and that this stimulation is fairly unspecific, primarily with respect to different anions. The response to cations is comparatively unspecific, too, with the exception of magnesium and calcium, which are known to interfere with biochemical processes.

As shown by Figs. 1, 3 and 4, the sudden burst of respiration, starting 1–2 min. after the addition of salt (concentrations of 0·01 up to 0 2 *M* were tested), depends upon a corresponding increase in the activity of the enzyme system. Cytochrome *c* as well as diphosphopyridine nucleotide show a sudden increase of the velocity of reduction in nitrogen and re-oxidation in air (measured as the reciprocal of half-time). Similar observations were made regarding cytochromes a_3, *a* and *b*, of which a_3 responds very rapidly. The lag at starting, that is, the time elapsing from switching on nitrogen to the start of

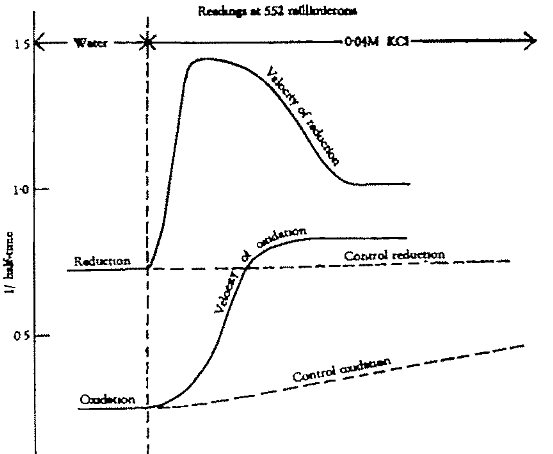

Fig. 3. Response of the velocities of reduction and re-oxidation to the addition of salt. Control and sample were both exposed to alternate gassing with nitrogen and air at constant intervals (see ref. 8) and the response at 552 mμ (cytochromes *c* + c_1) continuously recorded

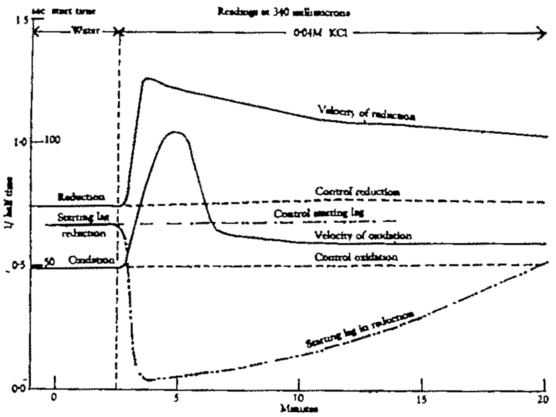

Fig. 4. Response of the velocities of reduction and re-oxidation and of the starting lag in reduction to an addition of salt. As in Fig. 3, but diphosphopyridine nucleotide at 340 mμ recorded

reduction (see ref. 8), drops considerably, too, a fact reflecting the rise in oxygen consumption; the dissolved oxygen disappears more rapidly if the cytochromes are activated.

It will be seen from Figs. 1, 3 and 4 that the sudden activation of the respiratory system attains an optimum a few minutes from the start and then slowly declines, even if the values still remain higher than in the controls. This fact may be explained as follows: the salt effect implies an acceleration of electrons through the respiratory system; this means a sudden over-consumption of substrate until a new balance between internal supply of substrate and oxygen consumption is attained.

Principle of Anion Respiration

The new experiments corroborate earlier results as to a non-specific salt (anion) effect[2-4]. This fact, as well as the observation that all enzymes in the respiratory chain are equally affected, is evidence against a closer combination between salt ions and enzymes. The new results are in full agreement with the principle of electrolytic regulation of the velocity of electron transference in the respiratory chain[2b]. It was originally assumed that electrons and anions are exchanged between single enzyme molecules (see below). This idea is still valid, but more recent results point to the existence of a spatially organized respiratory unit displaying electro-chemical properties similar to those of a galvanic cell.

Biochemical investigations are mostly confined to more or less purified and 'dissolved' enzymes, the reactions of which are proceeding in Newtonian media and obeying simple thermodynamic principles. It is, however, very unlikely that enzymes in the live protoplasm are behaving similarly to 'dissolved' enzymes. The regulated co-operation of numerous enzymes in the protoplasm points to the existence of a complicated spatial organization, in which the single enzymes occupy distinct areas. The activity of such enzymes will be guided by the spatial orientation of the molecules. It may be justified to speak of spatial biochemistry. The frequently used term 'multi-enzymes'[9] does not quite cover the conception of spatial organization, because important building stones of an enzyme system are 'factors' such as lipides, quinones, certain vitamins, etc. These factors serve as pathways or switchboards regulating the passage of electrons and intermediates between the reaction centres.

Little is known yet of reactions going on in organized enzyme systems, but it is possible to trace indirectly the existence of a spatial organization.

Observations of rhythmical pulsations in the stream of electrons through the respiratory system may be interpreted as a reflexion of its spatial organization[8,10]. Observations on homogenates of tissues point in the same direction. It was shown that even a high degree of mechanical disintegration of plant protoplasts does not change the quantitative relations between cytochromes a_3 and b. Spatially organized 'respiratory units' apparently compose the sub-micro-skeleton of the protoplasm, and it is reasonable to assume that these units are joined to microscopic structures, for example, surface layers of 'mitochondria'[12,4], the tonoplast of vacuolized cells, etc. We are now prepared to discuss the main problem, namely, the electrochemical properties of the respiratory unit.

According to recent observations[8,10], the respiratory system is composed of three spatially arranged groups of enzymes, namely: (1) enzymes linked with diphosphopyridine nucleotide; (2) flavoproteins and cytochrome b; and (3) cytochromes c and a_3. A number of accessory cytochromes (a, c_1, b_3, etc.) and 'factors' are additional elements of which some probably trap energy as high-energy phosphates. We know that diphosphopyridine receives electrons and protons from metabolites (*via* the tricarboxylic acid cycle) and transfers the electrons to the group flavoproteins + cytochrome b, which in its turn shuffles electrons to the group cytochromes $c + a_3$. From cytochrome a_3 the electrons leave the system with oxygen as the final acceptor.

The extremely rapid re-oxidation of a reduced respiratory unit indicates a location of the 'pole of oxidation' (o), namely cytochrome a_3, close to the surface of respiratory particles or membranes, whereas the 'pole of reduction' (i) namely, diphosphopyridine nucleotide, etc., is probably turned in a more or less opposite direction. As shown by Fig. 5, a stream of electrons moving from the pole of oxidation to the pole of reduction will then act as the internal circuit of a galvanic battery.

In the absence of an outer circuit, no electrons flow through a galvanic cell. A current starts, however, if the cell is surrounded by a salt solution, because the electrophoretic movement of ions in the outer circuit is a measure of the velocity of internal flux of electrons. The intensity of the current is regulated by the concentration (activity) of the salt ions.

This scheme may be applied to the conditions prevailing in the living cell. In the absence of salts, the stream of electrons from cytochrome a_3 to the dissolved oxygen molecules is slowed down to a minimum and the level of reduction of the respiratory enzymes rises. Because hydrogen ions and hydroxyl ions, together with anions of organic acids, are produced by the activity of the system itself, some 'respiratory idling' still proceeds; but observations on plant tissues and yeast show that lack of salts cuts down respiration to at least 50–75 per cent. Renewed supply of salts rapidly speeds up the stream of electrons. Because the capacity of dehydrogenation at the pole of reduction (Fig. 5) is less susceptible to changes, the immediate effect is a rising oxidation of the respiratory enzymes.

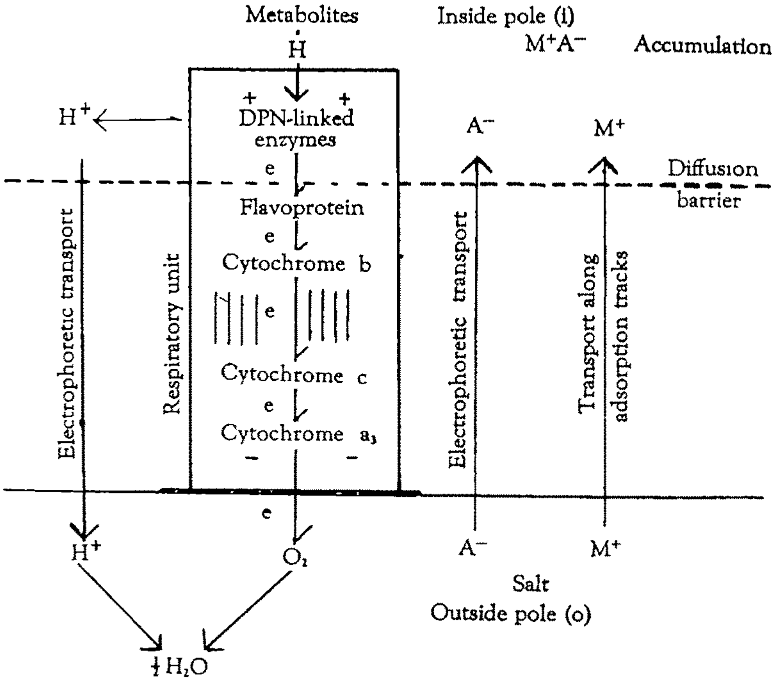

Fig. 5. Scheme of the principle of salt respiration. The respiratory unit is shown operating between surroundings containing free salts and an internal space behind a barrier which slows down the passive flow back of accumulated salts. The scheme may be applied to salt accumulation in the vacuoles of plant cells or in the interior of mitochondria. The initial salt absorption to carriers is omitted in this scheme (see text)

Biological Importance of Anion Respiration

The principle of anion respiration sheds light on a number of biological phenomena. It is a well-known fact that organisms produce electricity, even if only few of them, for example, the electric ray, store high-voltage static electricity. A circuit may be closed between the leaves and the roots of a plant and currents are also produced by root tips 1–2 cm. long[2b]. The respiratory character of the current is shown by the fact that the respiration rises or falls if the normal current is increased or diminished by means of an external supply of electricity. It may be expected that electric shocks directly affect the power of respiration, even if severe disturbances in the assembly of cations, which are adsorbed to cell structures, may be responsible for more 'explosive' effects.

Cytochrome respiration is a common condition for normal life and reproduction of cells, a thesis valid for almost all organisms—animals, plants and micro-organisms. A reflexion of the synthetic activity is the regulation of the amount of ion carriers referred to above. Malignant cells, for example, in cancer tumours, on the other hand, show a pronounced increase of glycolytic systems at the expense of cytochrome respiration[18]. Owing to stimulation of the cytochrome respiration, salts would perhaps help in curing tumours or mitigate deleterious effects of irradiation and/or to some extent protect normal cells from being affected by cancerogenous substances or irradiation. These are still unsolved problems. Deficiency in salts is a well-known cause of pronounced disturbances in the normal function of the animal organism.

Because transference of electrons occurs also outside of the scope of the 'respiratory unit' and because cytochromes may turn over electrons to acceptors other than oxygen, for example, in certain bacteria and in photosynthesis[15], salts may be able to display non-specific effects on many processes. An example of switching over glycolytic processes to a more efficient anaerobic utilization of glucose is given by wheat roots. In absence of oxygen, desalted roots show a 'glucose co-efficient' (consumed glucose/carbon dioxide produced) of 0·5. Addition of salt lowers the coefficient to values of about 0·33 or less.

The unspecific salt effect refers primarily to organized enzyme systems which are transferring electrons. It is, however, evident that a transference of electrons between two 'dissolved' compounds is also facilitated by salts. Salt anions will in this case probably be directly shuffled against electrons according to the scheme:

$$\text{reduced substance} + A^- \rightarrow (\text{oxidized substance}).A^- + e$$

provided that e is caught by an acceptor. Such a salt stimulation has been observed with regard to oxidation of cytochrome c (ref. 14).

According to Fig. 5 organized systems induce streaming of anions from the negative (o) to the positive (i) pole and of cations in the reversed direction. Because cations (H^+) are produced by the respiratory unit itself, but not anions, the regulating factor is primarily the supply of anions, hence the conception 'anion respiration'. If no semipermeable boundaries interfere, diffusion and mass streaming will perpetually counteract any appreciable accumulation of ions at the poles. The response of the respiratory units to salts will thus induce stirring in the surroundings and an indirect acceleration of the mixing of substances.

The synthesis of ion carriers is probably linked to the trapping of respiratory energy in products of phosphorylation[4], but very little is known about the chemical nature of these processes. The fact that cytochrome respiration maintains and reinforces the ability of the protoplasm to attract and retain salts is obviously of fundamental importance to the life of the cell.

The accumulation of free, that is, non-adsorbed, salts in the sap spaces of plant cells must be regarded as a special process implying the active transference of initially absorbed salts to the vacuoles. The retention of free salts is obviously caused by the operation of a barrier which precludes the rapid back flow of ions once accumulated. Fig. 5 illustrates the effect of a diffusion barrier placed between the poles of the respiratory unit. A monolayer of respiratory units will thus be able to accumulate anions. These anions will combine spontaneously with cations which are adsorbed to the protoplasm, as a consequence of which free salts are accumulated (see Fig. 5). It is feasible to assume that the vacuole membrance (tonoplast) acts in some such way. The fairly constant values of anion/oxygen observed in plant tissues point in this direction[3,4]. Accumulation of free salts would, of course, also occur in 'mitochondria' if the respiratory units are arranged in a monolayer coating the particles. Little is known, however, of true accumulation of salts in the 'free water' of protoplasts which do not contain vacuoles.

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[1] Lundegårdh, H., *Protoplasma*, 35, 548 (1941). Briggs, G. E., and Hope, A. B., *J. Exp. Bot.*, 9, 365 (1958).

[2] (*a*) Lundegårdh, H., and Burstrom, H., *Biochem. Z.*, 261, 235 (1933); 277, 223 (1935); (*b*) Lundegårdh, H., *Ann. Agric. Coll. (Uppsala)*, 8, 233 (1940); *Ark. f Bot. (Stockholm)*, 32 A, No. 12 (1945), *Ann. Agric. Coll. (Uppsala)*, 16, 339, 372 (1949). *Physiologia Plantarum*, 3, 103 (1950). "Encyclopedia of Plant Physiology" (Ruhland), 12, 2 (in the press) includes further literature.

[3] Robertson, R. N , *Aust. J. Exp. Biol. and Med. Sci.*, 22, 237 (1944).

[4] Lundegårdh, H., *Physiologia Plantarum*, 11, 332, 564, 585 (1958); 12, 336, 342 (1959).

[5] Lundegårdh, H , *Nature*, 167, 71 (1951); 169, 1088 (1952); 171, 521 (1953); *Ark. Kemi (Stockholm)*, 3, 69, 469 (1951); 5, 97 (1952); 7, 451 (1954)

[6] Bartley, W., and Davies, R. E., *Biochem. J.*, 57, 37 (1954).

[7] Lundegårdh, H., *Endeavour*, 18, 191 (1959).

[8] Lundegårdh, H., *Acta Chem. Scand.*, 10, 1083 (1956).

[9] Dixon, M , "Multi-Enzyme Systems" (1949). Neilands, J. B., Stumpf, P. K., and Stanier, R. Y., "Outlines of Enzyme Chemistry" (1955).

[10] Lundegårdh, H , *Biochim. Biophys. Acta*, 27, 653 (1958); 35, 340 (1959).

[11] Lundegårdh, H., *Biochim. Biophys Acta*, 27, 355 (1958).

[12] Robertson, R. N., *Endeavour*, 16, 193 (1957) Kling, H , *Protoplasma*, 49, 364 (1958).

[13] Warburg, O., "Schwermetalle als Wirkungsgruppen von Fermenten" (1946).

[14] Lundegårdh, H., *Nature*, 171, 477 (1953). Butler, G. W., *Physiologia Plantarum*, 6, 594 (1953). Boeri, E., and Tosi, L., *Arch. Biochem Biophys.*, 52, 83 (1954).

[15] Arnon, D. I., Allen, M. B., and Whatley, F. R., *Biochim Biophys. Acta*, 20, 449 (1956). Arnon, D I., *Nature*, 184, 10 (1959).

NEWS and VIEWS

New Year Honours List

THE following names of scientists and others associated with scientific work appear in the New Year Honours list:

Baron: Sir George Nelson, chairman of the English Electric Co., Ltd.

Baronet: Sir James Paterson Ross, president of the Royal College of Surgeons of England.

K.C.M.G.: Sir Charles Blackburn, chancellor of the University of Sydney, for services to education and medicine; Dr. George A. Currie, vice-chancellor of the University of New Zealand; Sir Philip Morris, lately chairman of the Commonwealth Education Conference, vice-chancellor of the University of Bristol; Geoffrey W. Nye, adviser on agriculture to the Secretary of State for the Colonies

Knights: Prof. Edward Ford, director of the School of Public Health and Tropical Medicine, University of Sydney; Dr. William H. Glanville, director of the Road Research Laboratory (Department of Scientific and Industrial Research); Dr John Hammond, formerly reader in agricultural physiology in the University of Cambridge; William W. S. Johnston, of the State of Victoria, for his work for postgraduate education in medicine; Prof. Harrie S. W. Massey, Quain professor of physics, University College, London.

C.B.: M. I. Michaels, under-secretary, Office of the Minister for Science; R. N. Quirk, under-secretary, Office of the Minister for Science; R. H. Weir, director-general of engine research and development, Ministry of Aviation; Dr. H. F. Willis, chief of the Royal Naval Scientific Service.

C.M.G.: A. W. Knight, chief commissioner of the Hydro-Electric Commission, State of Tasmania; R. E. Stedman, under-secretary, Ministry of Agriculture, Fisheries and Food; C. W. Michie, permanent secretary, Ministry of Agriculture, Northern Region, Nigeria; S. E. Morris, secretary for native affairs and chief native commissioner, Southern Rhodesia; Dr. E. W. Russell, director of the East African Agriculture and Forestry Research Organization.

C.B.E.: Prof. S. Alstead, regius professor of materia medica and therapeutics, University of Glasgow; H. E. Bannister, assistant secretary, Ministry of Agriculture, Fisheries and Food; J. D. Clark, director of the Rhodes–Livingstone Museum, Northern Rhodesia; C. E. Clarke, principal actuary, Government Actuary's Department; Dr. T F Davey, senior specialist (leprologist), Eastern Region, Nigeria; L. J. Davies, director of research and education, British Thomson–Houston Co., Ltd.; J. Edwards, head of Production Division and scientific adviser, Milk Marketing Board; Dr. A. J. P. Martin, for services in the development of chromatographic analysis; Dr. D. C Martin, assistant secretary of the Royal Society; Dr. A. T. R. Mattick, lately deputy chief scientific officer, National Institute for Research in Dairying, Shinfield, Reading; T. B. Nicol, engineer-in-chief, Metropolitan Water, Sewerage and Drainage Board, New South Wales; H. T. Pledge, keeper in the Science Museum, South Kensington; W. T. Price, principal of Harper Adams Agricultural College, Newport, Shropshire; H. T. Ramsay, director of the Safety in Mines Research Establishment, Ministry of Power; J. B. Randall, director of Veterinary Services and Animal Industry, Uganda; F. I. G. Rawlins, deputy keeper and scientific adviser, National Gallery; G. B. Ryle, director of forestry for England, Forestry Commission; Prof. R. W. Scarff, professor of pathology, University of London; V. C. Williams, of Griffith, New South Wales, for services to irrigation research.

From January 1960, issues of *NATURE* will be grouped into four volumes a year, instead of two as heretofore

Royal Society Vice-Presidents

THE president of the Royal Society, Sir Cyril Hinshelwood, has appointed the following vice-presidents for the year ending November 30, 1960: Sir William Penney, treasurer of the Royal Society, member for scientific research, United Kingdom

Atomic Energy Authority; Sir Lindor Brown, biological secretary of the Royal Society, Jodrell professor of physiology at University College, London; Sir William Hodge, physical secretary of the Royal Society, Lowndean professor of astronomy and geometry in the University of Cambridge; Dr. H. G. Thornton, foreign secretary of the Royal Society, lately head of the Department of Soil Microbiology at Rothamsted Experimental Station; Sir Patrick Linstead, rector of the Imperial College of Science and Technology; and Prof. Wilson Smith, professor of bacteriology at University College Hospital Medical School in the University of London.

Biology at Middlesex Hospital Medical School:

Prof. J. H. Woodger

PROF. J. H. WOODGER has just retired from the Middlesex Hospital Medical School after thirty-seven years as head of the Department of Biology. He graduated at University College, London, in 1914 and returned as assistant in zoology after four years active service, mainly in Mesopotamia, before accepting an appointment in the Middlesex Hospital Medical School at the age of twenty-eight. At a very early stage in his career, Woodger discerned that much of what passed for 'zoological philosophy' was a hotch-potch of confused thinking based upon an inattentive or insufficiently analytical use of words. In his "Biological Principles" of 1929 he did his utmost to dissect some of the stock antitheses of biology—for example, the antithesis between preformation and epigenesis—with the resources of ordinary language; but ordinary language proved too clumsy and imprecise, and Woodger's thoughts accordingly turned to the logical symbolism of Russell and Whitehead's "Principia Mathematica", enriched by the addition of many empirical constants for ideas which (unlike number or similarity) cannot be defined by logical concepts alone. His "Axiomatic Method in Biology" of 1937 was the first attempt to 'axiomatize' biology, that is, to cast it in the form of theorems logically deducible from a small number of axioms or postulates. Woodger has an international reputation as a logician; his "Technique of Theory Construction" of 1939 is a splendid exposition of the modern ideas on scientific methodology that arose mainly from the work of Karl Popper. Woodger's fine analytical work was continued in his Tarner Lectures of 1949–50 and is still actively in progress. His attempts at axiomatization have been thought premature by some who are competent to express an opinion; but the attempt to import rigour and precision into biological thinking was very much overdue, and Woodger's writing and thinking have had a quiet but deep and pervasive influence over a great deal of modern biological thought. Woodger stands in the mainstream of the great tradition of British empiricism; and his work marks the final repudiation of the metaphysical elements in *Naturphilosophie.*

Prof. D. R. Newth

PROF. WOODGER'S successor, Dr. D. R. Newth, is also a graduate of University College, London; and, like Woodger, he spent four years on active service between graduating in zoology in 1942 and returning to postgraduate work in 1946. Prof. Newth is an embryologist and the son of an embryologist; but narrow sectarian classifications are no longer appropriate in biology, and he is best described as a biologist of unusually wide knowledge and sympathies whose main concern has been with the biological problems raised by embryogenesis. His first work was on the normal development of amphibian embryos. He then turned to the largely neglected problems raised by the evolutionary history of the neural crest, and undertook an experimental analysis of its behaviour and fate in lampreys. After an interlude in which he worked upon the reactions of hagfishes to light, he returned to morphogenetic problems. His was the first rigorous attempt to answer the question: Can an animal 'regenerate' a part which it never possessed? In certain circumstances, he found, it can do so: by interfering with the primary embryogenesis of structures which are capable of regeneration in later life, they may develop abnormally; but these abnormalities can be partially made good if the abnormal part is amputated and so caused to grow anew. His analysis provides one possible approach to the problem of the persistence of post-embryonic life of the available genetic information in somatic cells Newth's appointment will give particular pleasure not only to biologists in Great Britain but also to his many colleagues on the editorial board of the international *Journal of Embryology and Experimental Morphology,* in the foundation of which he was one of the prime movers.

Forest Products Research Laboratory

THE Council for Scientific and Industrial Research has reviewed the operation of the arrangements made early in 1958, under which it asked the Timber Development Association to assume responsibility for certain aspects of timber research previously discharged by the Forest Products Research Laboratory. The Steering Committee for the Laboratory had become convinced that the new arrangements did not allow sufficient latitude to the Laboratory in the discharge of its essential duties, and that the division of functions between the Laboratory and any other research organization must be more flexible. It was sure, therefore, of the need for a new approach to the problem of securing industrial support of work done for the immediate benefit of the timber trade (which was the aim of the earlier arrangements) and also to provide for the needs of the users—the home producer (including the Forestry Commission), the merchant and the manufacturer. At the same time it was regarded as essential to maintain an adequate and well-balanced research team at the Forest Products Research Laboratory. The Research Council has therefore re-stated its general policy for the Forest Products Research Laboratory in the following terms: "There is a continuing need in the national interest, both for research of a fundamental character on timber (but with ultimate practical applications) and for work of immediate benefit to the Forestry Commission and timber users. This work is best done at the Forest Products Research Laboratory with financial support from industry wherever possible. The Laboratory's programme should be co-ordinated with that of the Timber Development Association Research and Development Committee and/or of any Research Association which may be set up, but should primarily be devised to ensure that the interests of Government and the whole timber industry (producers and users, as well as merchants) are served to the best advantage".

Graphite Blocks for the Dounreay Reactor

The Morgan Crucible Co., Ltd., of Battersea, London, S.W.11, has now been permitted to reveal that for the first time special borated graphite blocks were used by the United Kingdom Atomic Energy Authority in the assembly of the shield which surrounds the reactor vessel at the Authority's experimental station at Dounreay. The blocks were made and machined by the Morgan Crucible Co., at its works at Norton, near Worcester. This factory is not engaged in the production of nuclear pure material and was therefore chosen as one suitable for handling large quantities of boron compounds. Most of the borated graphite contained 0·3 per cent boron, but some contained 5 per cent—in both cases very uniformly distributed throughout the graphite. The blocks themselves were machined to five hundred different patterns, some of which were so intricate that wooden dummies were made to ensure that it was practicable to machine them in graphite. As a final check, the layers of finished graphite blocks were assembled at Norton. The blocks, painted with distinguishing coloured bands to avoid any risk of confusion with nuclear pure material, were then packed into cartons for the journey. Transport to Dounreay was by heavy lorry, which made one round trip of 1,500 miles per week, taking the whole dry season of one year—some eight months in all.

World Meteorological Organization Conferences and Symposia, 1960

The World Meteorological Organization has issued a provisional schedule of conferences and symposia to be organized in 1960: *January*, Meeting on Atomic Transport and Removal of Radioactive Debris from Nuclear Tests (New York); *March*, Facsimile Broadcasts of the Working Group on Telecommunications, established by Regional Association VI (Offenbach–Frankfurt); *April*, Eastern Caribbean Committee of Regional Association IV of North and Central America (Curaçao); *June* 27–*July* 15, Executive Committee—twelfth session; *August*, Working Group on Telecommunications of the Commission for Synoptic Meteorology; *August* 16–*September* 3, Commission for Maritime Meteorology—third session (Utrecht); *September–October*, Regional Association VI, Europe—third session (Lisbon or Madrid); South American Regional Seminar on Tropical Agrometeorology (Venezuela); Working Group on the Guide to Climatological Practice established by the Commission for Climatology (Geneva); *November* 21–26, Working Group on Telecommunications—established by Regional Association I, Africa—fifth session (Cairo); *November* 28–*December* 16, Regional Association I, Africa—third session (Cairo); *December* 1–16, Commission for Climatology—third session (London).

Recent Geophysical Work in Great Britain

The meeting of the Geological Society of London on January 27 will take the form of a symposium on "Geological Implications of Recent Geophysical Work in the British Isles". The afternoon session will discuss "The Study of Igneous Rock Masses". The principal speakers will be Dr. M. H. P. Bott (University of Durham), who will speak on granites, crustal structure and isostasy, and Dr. D. J. Blundell (University of Birmingham), whose subject will be the use of rock magnetism in studies of igneous geology. Other contributors will be Prof. T. Murphy (School of Cosmic Physics, Dublin) and Mr. S. H. U. Bowie (chief geologist, Atomic Energy Division, Geological Survey). The evening session will consider "The Study of Sedimentary Basins". The principal speakers at this session will be Mr. N. G. O'Halloran and Mr. R. G. W. Brunstrom (British Petroleum Co.), on the use of seismic reflexion methods in geophysical surveying, and Dr. W. Bullerwell (Geological Survey), on aeromagnetic surveys over central, eastern and southern England. Prof. W. B. R. King and Mr. G. Armstrong (chief geologist, National Coal Board) will also speak. At the close of the evening session the discussions at both sessions will be summarized and reviewed by Prof. J. H. Taylor (King's College, London).

"Inadequacy of Scatter Mechanisms in Tropospheric Radio Propagation"

In a communication under this title in *Nature* of November 14, p. 1558, it was stated that "the lenses have an average size of a few kilometres diameter" Mr. P. C. M. de Belatini states that this should read "a few metres". The toroids themselves are believed to be on average about 600 metres in diameter with an 'eye' that acts as the lens of about 15 metres diameter. These dimensions, of course, vary with time.

Announcements

Mr. B. A. Worswick, recently chief engineer of Fischer and Porter, Ltd., has been appointed technical director with a seat on the Board of Elcontrol, Ltd. He was previously chief development engineer of Bailey Meters and Controls, Ltd., after having served for some years with Imperial Chemical Industries, Ltd.

The International Sericultural Commission has announced that the second International Technical Sericultural Conference will be held in Murcia, Spain, during April 2–5. Further information can be obtained from A. Schank, General Secretary-founder, Commission Séricicole Internationale, Station de Recherches Séricicoles, 28 Quai Boissier de Sauvages, Alès, France.

The Brighton Technical College School of Pharmacy has announced that a course of eight evening lectures on Spectroscopic Methods of Pharmaceutical Analysis will be held during the period February 9–March 29, at the Main Hall of the College, Richmond Terrace, Brighton 7. Further details can be obtained from Dr. J. C. Parkinson, School of Pharmacy, Technical College, Brighton 7.

A conference on the Actinomycins and their Importance in the Treatment of Tumours in Animals and Man will be held under the auspices of the New York Academy of Sciences at the Barbizon-Plaza Hotel in New York City during March 31–April 1. Further information can be obtained from the Executive Director, New York Academy of Sciences, 2 East 63rd Street, New York 21, N.Y.

Errata. In the communication entitled "A Functional Interpretation of the Electron-Microscopic Structure of the Sensory Hairs in the Cristæ of the Elasmobranch *Raja clavata* in Terms of Directional Sensitivity" in *Nature* of December 5, p. 1807, Prof. O. Lowenstein points out that there are two mistakes: par. 3, line 1, for "when" read "why"; a grant acknowledged in the penultimate paragraph was from the Swedish Medical Research Council, not the Karolinska Institutet as printed.

CORNELL UNIVERSITY CENTER FOR RADIOPHYSICS AND SPACE RESEARCH

A NEW research centre is to be set up by Cornell University, Ithaca, New York, for the purpose of conducting research, and training research workers, in the subjects of radiophysics and space research. The centre will operate a high-powered radar equipment in Puerto Rico, financed by the Department of Defense. This equipment will make use of a fixed parabolic mirror with a diameter of 1,000 ft. built into a natural hollow in the mountains. It will have a peak power of 2·5 MW. In addition, it is proposed that there should be a radio-astronomy receiver near Ithaca, and a transmitter on the campus of Cornell.

The director of the new Centre will be Prof. T. Gold, who will be assisted by Prof. H. G. Booker as associate director. Both Gold and Booker went to the United States from Great Britain. Both worked on radar in England during the Second World War, Gold at the Admiralty Research Establishment, and Booker at the Telecommunications Research Establishment of the Ministry of Aircraft Production. After the War, Gold became a Fellow of Trinity College, Cambridge, and a demonstrator in the Cavendish Laboratory. Later he joined the Royal Observatory, Greenwich, as a chief assistant, and in 1957 he left for America, where he became professor of astronomy at Harvard. He is noted for the originality of his researches in astronomy, geophysics and the theory of hearing.

Booker took the Mathematical Tripos at Cambridge in 1933 and afterwards worked on the theory of electromagnetic waves in close collaboration with the radio section of the Cavendish Laboratory. He became a Fellow of Christ's College and a University lecturer in mathematics. During the War he led the theoretical division at the Telecommunications Research Establishment and was responsible for originating the 'duct' theory of super-refraction of radio waves. He returned to Cambridge after the War and played an important part in developing the theory of the scattering of radio waves from ionospheric irregularities. He left Cambridge for the United States in 1948, and became professor of electrical engineering at Cornell University. There he extended the theory of scattering to include the effect of irregularities in the troposphere, and he was one of the leaders in the team which turned this scattering to practical use in commercial 'forward scatter' radio links. He will retain his present position at Cornell in addition to becoming associate director of the new Center.

Both Booker and Gold are noted as mathematicians who can not only talk the language of the physicists but appear also to think in that language. They seem to have an intuitive grasp of fundamental principles which enables them to see at once what a piece of mathematics really implies. Together with the physicists, engineers and mathematicians at Cornell, all of whom will be called upon to help them from time to time, they will form a powerful team.

In one set of investigations the high-power radar equipment will be used to observe radio-echoes from the planets. In another it will be used for the observations of incoherent back-scatter from the electrons in the terrestrial ionosphere. Its use for the latter purpose will follow a suggestion by Gordon, which has already been tried out successfully by Bowles, that incoherent scattering might be used to investigate the upper side of the ionosphere. Gordon is to be a member of the Cornell team at the new Centre.

All radio- and space-physicists will look forward to valuable results from the new Center.

J. A. RATCLIFFE

RESEARCH BY McGILL UNIVERSITY IN BARBADOS

Bellairs Research Institute

THE Bellairs Research Institute of McGill University was founded in 1954 by the late Commander C. W. Bellairs, R.N., for the purpose of providing the University with research facilities in tropical biology. The first laboratory was put into operation in the seaside residence of Commander Bellairs in 1955. An increasing interest in the Caribbean area on the part of McGill soon made it apparent that the original quarters were not adequate for an expanding research programme. Plans were drawn up for a new building which was completed early in 1959.

While the present Institute quarters were designed primarily for the pursuit of studies in marine biology, the needs of workers in other fields of biology have not been overlooked in the construction of the building. The laboratory consists of three bright and airy work rooms with water tables and running fresh water and sea-water. There are a large concrete aquarium and outdoor tubs for the storage of freshly caught material. There are also an office, store-rooms, photographic room and microscope room. Outlets for bottled gas have been installed and the electric supply is 110 volts, 50 cycles. Accommodation, consisting of single and double bedrooms, lounge and kitchen, is available for a limited number of visiting investigators.

There are several small boats with outboard motors for collecting in shallow water and a 30-ft. diesel launch for work at sea. The launch is equipped with a winch and gear for oceanographic and plankton collections. Diving equipment, including aqualungs, an underwater camera and a variety of nets, dredges and other collecting gear, is available.

The Institute is situated on the western coast of Barbados, some seven miles from the capital, Bridgetown. A well-developed coral reef lies directly in front of the laboratory and extensive reefs lie along most of the western side. Around the rest of the island there is a wide variety of marine environments, including rocky shores and shallow-water flats of sand, mud or grass. Along the southern coast there are extensive shallow banks of corals and gorgonids, and many deep-water forms such as crinoids and antipatharians are found in 30–40 ft. of water. Deep oceanic water comes to within a mile or two of shore and the island lies close to the convergence of the North and South Equatorial currents.

Tropical Research Laboratory

Operating now in its second year is the McGill University Tropical Research Laboratory, located at Waterford, Barbados. The laboratory, which is administered through the departments of Geography and Meteorology of McGill University, has undertaken, in its initial stage, investigations into climatology and agricultural meteorology in the Caribbean and plans later to expand into a meteorological station as well as a graduate centre in the tropics for geographical research.

Among its immediate activities is the measurement of potential and latent evaporation at a tropical station. These results, with the usual meteorological data, will be published at regular intervals each year for the information of interested research workers in the field of climatology. Results of these investigations will also be used locally in experiments planned to aid the Department of Agriculture in the expansion of irrigation and land-use schemes.

PEST INFESTATION RESEARCH, 1958

THE report of the Pest Infestation Research Board of the Department of Scientific and Industrial Research for 1958* reflects the increased concern with problems of food storage and handling in overseas territories and on board ships, and with the problems that may arise when grain is stored for very long periods under different conditions of temperature and of oxygen concentration. Two long-term experiments have been set going, in which the effect of low temperature (4·5 ± 0·5 °C.) and low oxygen concentration (between 1 and 2 per cent), singly or in combination, on the keeping quality of dry wheat, will be investigated. A hard Canadian and a soft English wheat are being used. Chemical analyses, milling tests and bread-making tests were made initially and will be repeated at intervals over a period of ten or twelve years. Since it is quite inexpensive to maintain large stores of grain in these conditions the outcome of this investigation will be of considerable practical importance.

Films of insecticidal lacquers containing dieldrin and endrin have proved very effective in killing adult flour beetles (*Tribolium confusum*). It has now been shown that flour up to at least 2 in. deep becomes toxic to the beetles after being in contact with the lacquer for two weeks and retains this toxicity for many months. Until the amount of contamination that might occur in practice has been established, caution in the use of these particular insecticidal preparations is advocated. Whereas much is known about the susceptibility to insecticides of the insects of stored products, very little is known about the mites. An investigation of the flour mite (*Acarus siro*) is now being undertaken to make good this deficiency.

The use of gas-proof sheets for covering produce during fumigation has been increasing greatly in recent years. But users have been unable to obtain guidance on the selection of the material best suited to this purpose. Some have proved unsuitable because of lack of durability of the coating, others are highly permeable to methyl bromide and yet others become stiff after contact with liquid methyl bromide. A systematic study has now been carried out in co-operation with manufacturers, and the Laboratory should soon be in a position to put forward agreed specifications.

In addition to these problems of immediate practical importance the Laboratory has continued to produce a large amount of basic information on the insects and other organisms affecting stored products. This has consisted in detailed life-history studies in controlled physical environments, the systematics of insect pests, the response of insects to fumigants and other insecticides. In the Biochemistry Section work of even wider interest has been carried out. The physiological and toxicological studies on the mode of action of insecticides and the biochemical nature of acquired resistance to insecticides being carried out in that section is fundamental to applied entomology of every kind.

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* Department of Scientific and Industrial Research. Pest Infestation Research: The Report of the Pest Infestation Research Board with the Report of the Director of Pest Infestation Research. Pp v+55+8 plates. (London: H.M. Stationery Office, 1959.) 5s. net.

X-RAY MEASUREMENT OF THERMAL EXPANSION PERPENDICULAR TO THE LAYER PLANES OF ARTIFICIAL AND NATURAL GRAPHITES

By E. G. STEWARD and B. P. COOK
Research Laboratories, The General Electric Co., Ltd., Wembley

THERE are apparently conflicting data in the literature concerning X-ray diffraction measurements of (*a*) the thermal expansion of graphitic carbons and (*b*) the possible variation of the expansion coefficient with the proportion (p) of disordered (turbostratic) stacking of the layer planes.

For natural (Ceylon) graphite (p almost zero) over the range 15–800° C., the variation of the interlayer spacing d (A.) with temperature t (deg. C.) is given by Nelson and Riley[1] as $d = 3 \cdot 3525 + 90 \cdot 54 \times 10^{-6}\, t + 6 \cdot 33 \times 10^{-9}\, t^2$ (giving an average expansion coefficient (α) defined as $\frac{d_2 - d_1}{d_1 \Delta t}$, of 28·5 ± 0·1° C.$^{-1}$) and by Matuyama[2] as $d = 3 \cdot 3535 + 82 \cdot 41 \times 10^{-6}\, t + 10 \cdot 30 \times 10^{-9}\, t^2$ ($\alpha = 27 \cdot 0 \pm c.\ 0 \cdot 1°$ C.$^{-1}$) (Fig. 1). If in either determination the experimental errors have been so under-estimated that the difference in the resulting values of the average expansion coefficient is not in fact significant, then the inclusion in the appropriate

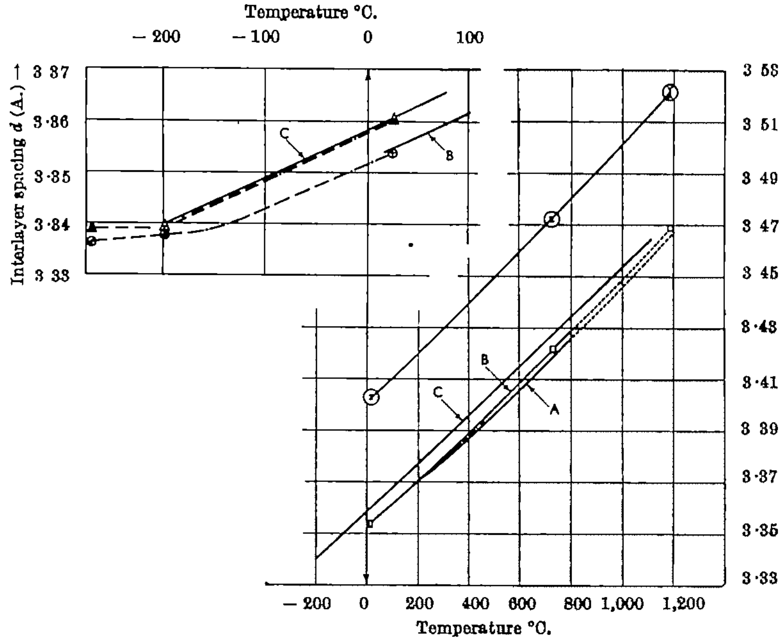

Fig. 1. Interlayer spacings of graphitic carbons as a function of temperature. *A*, $p \simeq 0$ (Matuyama); *B*, $p \simeq 0$ (Nelson and Riley); *C*, $p \simeq 0{\cdot}2$ (Walker, McKinstry and Wright); ⊕, $p \simeq 0$ (Baskin and Meyer); ▲, $p \simeq 0{\cdot}2$ (Baskin and Meyer); – – –, extrapolations to 1,200° C. from given equations; □, denotes present results and error limits for $p \simeq 0$; I, denotes present results and error limits for $p \simeq 0{\cdot}75$; – · – · –, see text

equation of the small term in t^3 would not be justifiable.

For an artificial graphite with about 20 per cent disordered stacking ($p \simeq 0{\cdot}2$) over the range –196–1,118° C., Walker, McKinstry and Wright[3] obtain $d = 3{\cdot}358 + 95{\cdot}2 \times 10^{-6}\, t$ (Fig. 1) with a standard deviation from linearity significantly smaller than the above βt^3 terms at high temperature. From their results Walker *et al.* calculate a decrease of 13 per cent in interlayer binding-energy between room temperature and 1,118° C. From a comparison of their results with those of Nelson *et al.* for the more perfect graphite, they conclude that the increase in d for a given rise in temperature is independent of p. By using the known[4] relationship between d and p one could then deduce that the expansion is independent of d—as found experimentally by Walker for their own artificial graphite.

Some recent qualitative work by Matuyama[5] on a range of essentially non-graphitic carbons is inconclusive on the issues under discussion.

Baskin and Meyer[6] report considerable differences in the amounts of crystal structure contraction of artificial ($p \simeq 0{\cdot}2$) and natural ($p \simeq 0$) graphite on cooling from room temperature to 4·2° K. (Fig. 1). Their 'average' expansion coefficient (based on two points 78° K. and 297° K.) for the artificial graphite agrees well with Walker's value but for natural graphite is lower than Nelson and Riley's value over 15–800° C. They suggest that the smaller changes they observe in cooling from 78° K. to 4·2° K. result from anisotropic contraction of crystallites producing mutually restraining stresses; these would be greater for artificial graphites having smaller crystals than for natural graphite.

To minimize dependence on absolute accuracy in making comparisons between different types of graphite, we have made measurements in the same apparatus under the same conditions, on both a natural graphite ($p \simeq 0$) and on a coke. To enhance any differences dependent on crystal structure stacking disorder, a coke with a large proportion ($p \simeq 0{\cdot}75$) of turbostratic structure was chosen. Results for room temperature, 728° C. and 1,188° C. (actual values (± 6° C.) determined by platinum lattice expansion calibration), are shown in Fig. 1. For the natural graphite where all reflexions arise from ordered structure, $000l$ and $11\bar{2}l$ reflexions were used and correction was made in the conventional way for absorption, etc. For the less graphitic material, although full correction to put the results on an accurate absolute scale was not possible, relative changes in 0004 reflexion angles were measured with sufficient precision for this initial survey. Thus, while the gradient for $p \simeq 0{\cdot}75$ has an accuracy limited only by the relative errors (indicated in Fig. 1) for the individual points, the numerical values for any temperature probably have a slightly greater error (± 0·005 A.). In the range 15–728° C. for $p \simeq 0$, $\alpha = 28{\cdot}4 \pm 0{\cdot}7°\ \text{C.}^{-1}$ and for $p \simeq 0{\cdot}75$, $\alpha = 29{\cdot}2 \pm 0{\cdot}7°\ \text{C.}^{-1}$.

Several significant features may be noted from the present results and survey.

The new measurements on the natural graphite closely follow the earlier findings of Nelson and Riley and give an indication at 1,188° C. of slight departure from linearity. For our less-graphitic material ($p \simeq 0{\cdot}75$) there is again approximate linearity and the expansion coefficient over the range 15–1,188° C. is the same within present limits of accuracy as that for the fully graphitic carbon. Thus in the range of interlayer spacings involved and over this temperature-range the coefficient of expansion perpendicular to the layer planes appears to be largely independent of d and p. The evidence of some curvature for the most perfect graphites supports the view that the coefficient of expansion must, however, be a function of temperature: further investigation at higher temperatures is clearly desirable.

For fully ordered graphite at 78° K. and 4·2° K., rather than deduce with Baskin and Meyer that the expansion coefficient is "distinctly lower", it is suggested that the variation of d with t is following the course shown thus –·–·– in Fig. 1; at these lower temperatures contraction is eventually limited by the repulsion forces between adjacent layer planes. Supporting this view is the observation that for the artificial graphite with $p \simeq 0{\cdot}2$ and correspondingly larger d, Walker's results show uninterrupted and essentially linear contraction from higher temperatures down to 78° K. At the lowest temperatures, where for either material the interlayer spacing is becoming critical, thermal contraction becomes smaller. Presence of imperfections in the structure also becomes important, and this may well explain the different ultimate behaviour reported by Baskin and Meyer between their materials: in particular, it would be expected that turbostratic stacking would have a larger limiting *d*, since the random atomic arrangement between adjacent layers will hinder their close approach.

It is hoped to report and discuss these results in more detail elsewhere and when further investigations at high and low temperature have been made.

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[1] Nelson, J. B., and Riley, D. P., *Proc. Phys. Soc.*, **57**, 477 (1945).
[2] Matuyama, E., *Tanso*, **7**, 12 (1958).
[3] Walker, P. L., McKinstry, H. A., and Wright, C. C., *Indust. Eng. Chem.*, **45**, 1711 (1953).
[4] Franklin, R. E., *Acta Cryst.*, **4**, 253 (1951).
[5] Matuyama, E., *Nature*, **184**, 544 (1959).
[6] Baskin, Y., and Meyer, L., *Phys. Rev.*, **100**, 544 (1955).

PLANT THIOGLYCOSIDES AND THE PROBLEM OF ENDEMIC GOITRE IN AUSTRALIA

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CLEMENTS and Wishart[1] have suggested that an appreciable amount of endemic goitre in certain areas of Tasmania may be due to a goitrogen present in the milk of cows fed 'chou moellier'. Apart from other evidence, milk and extracts of skim milk from chou moellier-fed cows were found to depress the uptake of iodine-131 by the thyroid glands of experimental animals and human volunteers. Samples of milk from the goitrous area of Warwick (Queensland), where in spring and early summer the pastures are heavily contaminated with turnip weed (*Rapistrum rugosum*), were also shown to be goitrogenic[2]. By implication, other cruciferous weeds might be considered suspect in relationship to the general hypothesis of an association between plant thioglycosides and endemic goitre in certain circumstances.

Greene, Farran and Glascock[3], in comparing milk from cows fed hay, chou moellier or pastures lightly contaminated with cruciferous weeds, found the latter to have the highest goitrogenic activity. Factors complicating the comparison of the results of investigators in different parts of the world could be alterations in the content of thioglycosides in the plant due to differences in variety, soil and climatic conditions, or modifications of the rumen flora. On the other hand, Virtanen, Kreula and Kiesvaara[4] examined the milk of a cow 12 hr. after it had been fed moistened rape seeds (equivalent to 4·5 gm. (–)-5-vinyl-2-oxazolidinethione (goitrin[5])) and could detect only a small amount of a substance showing a similar spectral absorption curve to goitrin but with an absorption maximum at 255 mμ instead of 240 mμ.

In association with the Goitre Sub-Committee of the National Health and Medical Research Council, we have been engaged on a systematic investigation of the thioglycosides—and their aglycones—of chou moellier and of those cruciferous weeds which are prevalent as contaminants of pastures in goitrous areas of Tasmania and southern Queensland. We wish to report here some of the results which we have obtained in a study of *Rapistrum rugosum*.

The seeds of this plant yielded unexpectedly only traces of thioglycosides, whereas chromatographic examination of extracts of the mature fruit, including seeds, revealed the presence of one major and one minor component. From such extracts, after treatment with myrosin, we were able to separate the major aglycone in crystalline form (m.p. 45° C.; 1·2 gm. per kgm. dry weight) and to establish its identity as γ-methylsulphonyl-propyl-*iso*thiocyanate, a substance previously isolated from wallflower seeds (*Cheiranthus cheiri*) by Schneider[6] and named by him 'cheiroline'. Schneider was able to confirm the structure of cheiroline by synthesis (see also Kjaer, Marcus and Conti[7]. We are indebted to Prof. Kjaer for an authentic sample of cheiroline).

Cheiroline was also isolated from the tissues of *Rapistrum rugosum*—in the case of extracts of fresh leaves, 0·4 gm. crystalline aglycone per kgm. wet weight. The yields of pure aglycone from fruit and leaves, which must be regarded as minimum values, would indicate a potentially high daily intake by a cow grazing on heavily contaminated pastures.

The results of experiments to compare the action of cheiroline with that of known goitrogens are summarized in Table 1.

Table 1. COMPARISON OF EFFECTS OF CHEIROLINE, THIOURACIL AND GOITRIN ON IODINE-131 UPTAKE (1 μC.) BY THE THYROIDS OF 100 GM. MALE RATS

Expt. No.	Mean net counts per min.				Per cent control group		
	Control	Cheiroline	Thiouracil	Goitrin	Cheiroline	Thiouracil	Goitrin
1*	23,710	3,700	2,178	—	16	10	—
2*	14,893	4,339	1,710	—	29	11	—
3†	10,313	5,474	—	885	55	—	9
4†	13,369	5,071	—	1,170	40	—	9·5

Dosage: cheiroline, 5 mgm.; goitrin, 5 mgm.; thiouracil, 1 mgm.; each in 1 ml. saline. Control, 1 ml. saline. Four animals per group.
* Mixed strain, Wistar and *Rattus norvegicus*.
† Wistar strain.

The substances were injected subcutaneously 1 hr. prior to the intraperitoneal injection of iodine-131, the animals killed 3 hr. later and the thyroids dissected, weighed and digested overnight and then counted in a scintillation counter (compare ref. 8). On the basis of this test, cheiroline is goitrogenic, although the degree of response varied, with the differences most marked between the two strains of animals used. It was considered unlikely that variation in iodine intake was a factor, as the dietary pattern in our colony (Wistar strain) was very similar to that of the mixed strain obtained from another source in Melbourne, but that possibly the *iso*thiocyanate group had been modified *in vivo*, thereby introducing another variable. By contrast, the activities of goitrin and thiouracil, although measured in different strains, had approximately the same ratio (1 : 6) as that recorded by Astwood, Greer and Ettlinger[5]. According to Kjaer[9], *iso*thiocyanates are inactive or only weakly active as goitrogenic agents (see also Wagner-Jauregg and Koch[10]).

Since we have a prime interest in the behaviour, in the rumen, of cheiroline (and other thioglycoside aglycones), we examined its stability in this medium. Cheiroline was incubated with freshly taken rumen liquor, after filtration through butter muslin, as set out in Table 2.

Table 2. INCUBATION OF CHEIROLINE WITH RUMEN LIQUOR

	Blank	Controls		Treated	
		A	*B*	*A*	*B*
* Rumen liquor (ml.)	25	25	25	25	25
Buffer (0·1 *M* PO_4; *p*H 6·4)	10	10	10	10	10
Urea (mgm.)	—	60	60	60	60
Cheiroline (mgm.)	—	—	—	50	50
Incubation period (hr.)	—	2	5	2	5
† Final *p*H	—	6·8	6·9	6·8	6·9
‡ Ammonia (μmole/ml.)	13	33·5	39	25·5	38

* *p*H 6·4.
† Initial *p*H assumed in all cases as for blank (6·35).
‡ As volatilized from NaOH-treated samples.

Urea was added to produce levels of ammonium ion comparable to those attained in animals grazing during the flush spring growth. At the conclusion of the incubation period, the contents of the bottles were brought to 85° C., centrifuged, and the supernatants dried in the frozen state. When samples of chloroform extracts of the residues were applied to paper and run in chloroform/water and then sprayed with Grote's reagent[11] or ammoniacal silver nitrate solution, the control chromatograms were negative, whereas, on those corresponding to the flasks to which cheiroline had been added, this substance was absent, but there was a strong spot at R_F 0·12, with a minor spot which ran at the same R_F as cheiroline monothiourea (close to the origin). There was no difference between the chromatograms corresponding to the 2- and 5-hr. incubation periods. The combined chloroform extracts were passed twice through an alumina column with 80 per cent ethanol used each time to elute a slightly coloured upper band. The oily residue from the second eluate was washed several times with small amounts of ether and chloroform, and it was then obtained crystalline from methanol (yield about 12 mgm.). The substance slowly developed a permanent bluish green colour in dilute acetic acid with the addition of potassium ferrocyanide[12], as did also cheiroline monothiourea but not cheiroline itself. It was recrystallized from methanol (m.p. 117–119° C.), analysed (found: S = 30·5 per cent) and identified as *sym.* di-cheiroline thiourea ($(CH_3SO_2CH_2CH_2CH_2NH)_2C{=}S$), when compared with a synthetic sample, on the basis of R_F values—in chloroform/water (Fig. 1) and *n*-butanol/ethanol/water, 4 : 1 : 4—analysis (calc.: S = 30·3 per cent) and mixed melting point (which was undepressed). This compound separated rapidly in high yield when equimolecular proportions of cheiroline and γ-methylsulphonyl-propylamine hydrochloride were mixed in ethanolic solution in the presence of triethylamine. The synthetic material was recrystallized from methanol (m.p. 121–122° C.; Schneider[6] reported 125–126° C. (from water)).

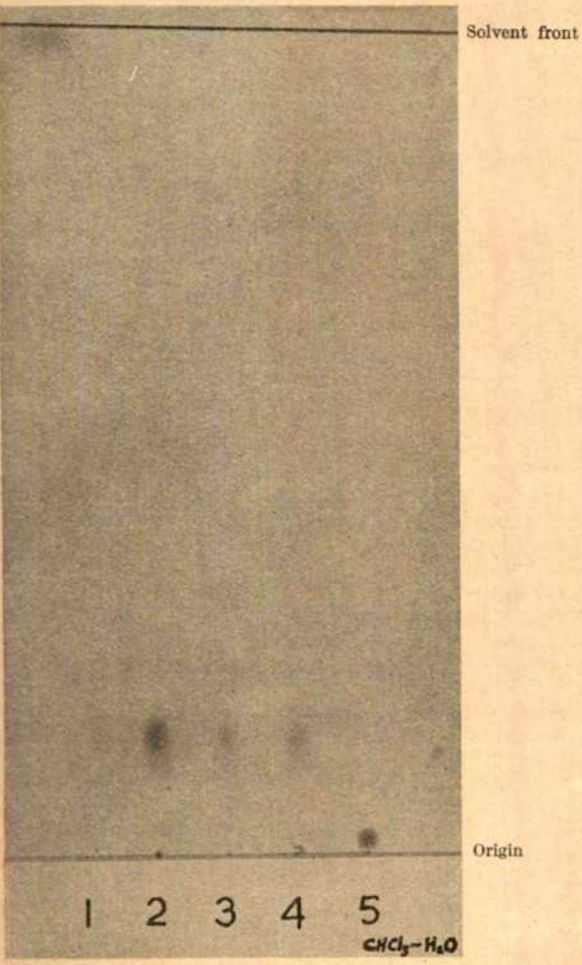

Fig. 1. (1) Cheiroline; (2) crude chloroform extract; (3) synthetic di-cheiroline thiourea; (4) crystalline substance from chloroform extract; (5) cheiroline monothiourea. Cheiroline running with solvent front. Chromatogram sprayed with ammoniacal silver nitrate

A few disubstituted thioureas have been shown to be goitrogenic in the rat[13,14] and, in the case of dibenzylthiourea, in the rabbit[15] (by mouth but not when injected). Diethyl thiourea by mouth was found by Astwood[13] to be a more active goitrogen in the rat than thiourea. Ethyl *iso*thiocyanate is one of the numerous aglycones of this type recorded as occurring in Nature[16,17], very few of which possess a hydroxyl group in the appropriate position for spontaneous cyclization to derivatives of 2-oxazolidinethione related to goitrin. It may be noted that, although we have isolated crystalline goitrin from chou moellier seeds in amount equivalent to 2 gm. per kgm., this substance corresponds to but one of four major thioglycoside fractions present in the seed extracts.

In preliminary tests using the above-mentioned rat assay method, di-cheiroline thiourea proved to be only weakly goitrogenic (iodine-131 uptake 70 per cent of the control (dose 5 mgm.) compared with cheiroline 30 per cent (5 mgm.) and thiouracil 12 per cent (1 mgm.)). The results following other routes of administration may well be different, since the thiourea, when pure, is practically insoluble in ether and chloroform, sparingly so in methanol and appreciably soluble in water, properties which are largely the reverse of those shown by cheiroline. There are, moreover, several cases where the rat assay has been shown to be an unreliable index of goitrogenic potency in man, for example, of 1-methyl-2-mercaptoimidazole[18].

We are at present more interested in the questions as to whether the transformation of *iso*thiocyanates in rumen liquor is a general reaction, whether it occurs in the rumen *in vivo*, and, if so, whether the products can be absorbed and transferred to milk.

There are thus certain obvious extensions of the experiments reported here—and several of these are in progress—before a general concept of the transformation in the rumen of *iso*thiocyanates into goitrogenic disubstituted thioureas can be added to the hypothesis stated at the beginning of this article.

We are indebted to Dr. F. W. Clements for organizing the supply of suspected plants and their seeds; to Dr. M. T. McQuillan for her co-operation in the rat assays; to Dr. F. J. R. Hird for the ammonia values; and to the National Health and Medical Research Council for a personal grant (H. S. B.) and financial assistance towards the costs of the investigation.

[1] Clements, F. W., and Wishart, J. W., *Metabolism*, **5**, 623 (1956).
[2] Clements, F. W., *Med. J. Aust.*, **2**, 645 (1957).
[3] Greene, R., Farran, H., and Glascock, R. F., *J. Endocrinol.*, **17**, 272 (1958).
[4] Virtanen, A. I., Kreula, M., and Kiesvaara, M., *Acta Chem. Scand.*, **12**, 580 (1958).
[5] Astwood, E. B., Greer, M. A., and Ettlinger, M. G., *J. Biol. Chem.*, **181**, 121 (1949).
[6] Schneider, W., *Ann.*, **375**, 207 (1910).
[7] Kjaer, A., Marcus, F., and Conti, J., *Acta Chem. Scand.*, **7**, 1370 (1953).
[8] McGinty, D. A., Rawson, R. W., Fluharty, R. G., Wilson, M., Riddell, C., and Yee, H., *J. Pharmacol. Exp. Therap.*, **93**, 246 (1948).
[9] Jensen, K. A., Conti, J., and Kjaer, A., *Acta Chem. Scand.*, **7**, 1267 (1953).
[10] Wagner-Jauregg, T., and Koch, J., *Z. Naturf.*, B, **2**, 14 (1947).
[11] Grote, I. W., *J. Biol. Chem.*, **93**, 25 (1931).
[12] Sato, T., *Biochem. Z.*, **23**, 45 (1910).
[13] Astwood, E. B., *J. Pharmacol. Exp. Therap.*, **78**, 79 (1943).
[14] Stanley, M. M., and Astwood, E. B., *Endocrinol.*, **41**, 66 (1947).
[15] Hüter, F., *Z. Naturf.*, B, **2**, 19 (1947).
[16] Kjaer, A., and Larsen, I., *Acta Chem. Scand.*, **8**, 699 (1954).
[17] Kjaer, A., *Suomen Kemistilehti*, A, **32**, 53 (1959).
[18] Stanley, M. M., and Astwood, E. B., *Endocrinol.*, **44**, 588 (1949).

OCCURRENCE OF AN ORGAN-SPECIFIC ANTIGEN ASSOCIATED WITH THE MICROSOME FRACTION OF PLANT CELLS AND ITS POSSIBLE SIGNIFICANCE IN THE PROCESS OF CELLULAR DIFFERENTIATION

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THE microsome particles of cells have received considerable attention from biologists because they are widely believed to play a major part in protein synthesis. However, little has been done to determine their role in cellular differentiation, although several workers have postulated their probable importance in this fundamental process (most recently by Bonner[1]). New light has been thrown on this aspect of microsome physiology while employing the Ouchterlony serum–agar technique[2] to study the changes which occur in the antigens of the wheat coleoptile during growth and differentiation and the distribution of these antigens in the other organs of the wheat plant (*Triticum vulgare*).

The wheat coleoptile during growth undergoes a period of cell division followed by one of differentiation. Cell counts using the method of Brown and Rickless[3] established that cell division was completed when the coleoptile had been growing for 54 hr. after sowing at 25° C. (var. King II) and thereafter, until 96 hr., growth was associated with cellular differentiation which in the coleoptile is largely a process of cell elongation.

Using an adjuvant technique[4], an antiserum was raised in a rabbit against a phosphate buffered protein extract (M/50; *p*H 7·6) of three thousand coleoptiles, containing samples of both early and late phases of growth and differentiation. A phosphate-buffered agar plate was set up as shown in Fig. 1 with this antiserum added to the centre cup. Approximately 1 per cent protein extracts of coleoptiles of different ages from 18 to 96 hr. were pipetted in the outer ring of cups. The antigen mixtures and antibodies were allowed to diffuse towards each other in an incubator at 37° C. for 20 hr. Precipitin bands formed, and where they joined one another it was assumed that they corresponded to the same antigen.

From a study of the frequency and distribution of mitotic figures in histological sections, together with information obtained from the change in cell number during growth, it was estimated that at 18 hr. and 24 hr. after sowing about 85 per cent of the cells of the coleoptile were either potentially able to divide or were dividing, the remainder being mainly in the early stages of differentiation. At 30 hr. about 70 per cent were dividing or potentially able to divide, at 42 hr. 30 per cent, and at 72 hr. 0 per cent, when all were differentiating or had differentiated. In the extracts representing the high percentage of dividing or pre-dividing cells three precipitin bands predominated (Fig. 1), and as the proportion of differentiating cells increased so other bands assumed dominance. Thus it appears that as differentiation proceeds the

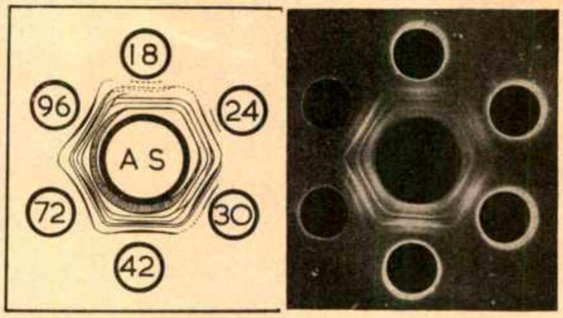

Fig. 1. Serological analyses of protein extracts representing different stages in the growth and differentiation of the wheat coleoptile. The figures in the outer ring of cups refer to the age of the coleoptile in hours from which the particular protein extract was made. The centre cup contained coleoptile antiserum possessing antibodies to all stages

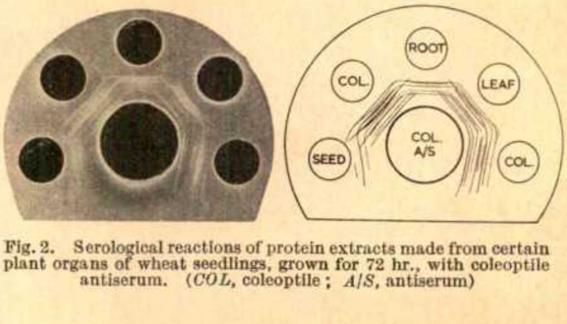

Fig. 2. Serological reactions of protein extracts made from certain plant organs of wheat seedlings, grown for 72 hr., with coleoptile antiserum. (*COL*, coleoptile; *A/S*, antiserum)

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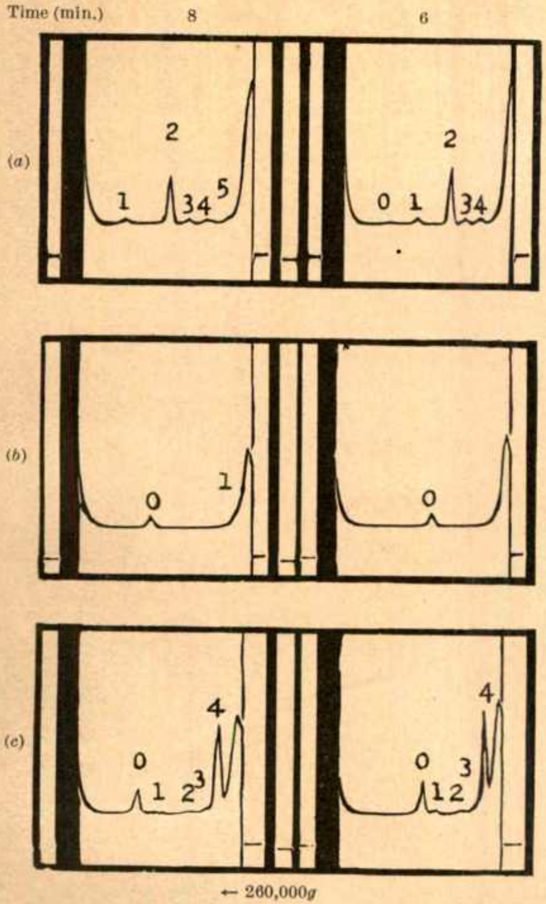

Fig. 3. Sedimentation diagrams of coleoptile, root and leaf extracts of wheat seedlings in 0·25 *M* sucrose, *M*/100 phosphate buffer *p*H 6·8. *a*, Dark-grown coleoptiles (48 hr. after sowing) ($S_{20,w} \times 10^{-13}$ values: peak 1 = 105; peak 2 = 69; peak 3 = 50; peak 4 = 33; peak 5 = 20). *b*, Roots (3 days after sowing) ($S_{20,w} \times 10^{-13}$ value: peak 0 = 73). *c*, Light-grown leaves (7 days after sowing) ($S_{20,w} \times 10^{-13}$ values: peak 0 = 74; peak 4 = 18)

principal differences in the cellular antigens are quantitative rather than qualitative. Nevertheless, it must be borne in mind that there could be qualitative antigenic differences existing between dividing and differentiating cells which were not apparent in these experiments because of the presence of a small proportion of differentiating cells in the extracts representing dividing cells.

To study the distribution of the coleoptile antigens throughout the plant, three-day-old wheat seedlings were dissected into coleoptiles, leaves, roots and seeds. These were extracted in phosphate buffer (*M*/20, *p*H 7·6) and, after centrifugation at 1,200*g* for 10 min., the extracts were pipetted into cups cut in a phosphate-buffered agar plate as shown in Fig. 2. The coleoptile antiserum was placed in the centre cup and the plate was incubated as described above. The coleoptile shared many of its precipitin lines with the leaf extract, as would be expected, since the coleoptile is in the nature of a modified leaf. The root and seed shared only a few precipitin lines with the coleoptile, although, doubtless, they possess their own specific antigens which were not detected with this antiserum.

Concurrently with these investigations an analytical ultracentrifugal study was carried out on coleoptile, leaf and root extracts. These extracts were made in phosphate-buffered (*M*/100; *p*H 6·8) 0·25 *M* sucrose and the results of the runs in the analytical ultracentrifuge are shown in Fig 3, *a*, *b* and *c*. All the extracts were found to contain a heavy component with a sedimentation constant within the range 69–74*S* (coleoptile = 69*S*; leaf = 74*S*; and root = 73*S*). Ts'o, Bonner and Vinograd[5] have reported that extracts of pea seedlings, after removal of the very large particles, contain a major component of 74*S*, which on analysis was found to contain 30–37 per cent ribonucleic acid, 55 per cent protein and 4·5 per cent phospholipids. They concluded that in size, chemical composition and morphology the particles resemble the nucleoproteins of the microsomes from animal tissues. Moreover, the 74*S* particle was found to dissociate at a *p*H of 8·5 into two particles of 58*S* and 38*S*, thereby forming a polymeric series[6]. Extracts were therefore made of coleoptiles (48 hr. after sowing) in 0·25 *M* sucrose at *p*H 6·8 and 8·4 (*M*/100 phosphate buffer) and it was found that the 69*S* component similarly dissociated at the higher *p*H and that the resulting $S_{20,w} \times 10^{-13}$ values of 69,50 and 33 corresponded to a polymeric series related as trimer, dimer and monomer, respectively (Fig. 4).

In order to determine whether any of the precipitin lines were associated with a 'microsome' antigen, the heavy components (mitochondria, nuclei, etc.) were first removed by a preliminary centrifugation at 10,000*g* for 30 min. from a 0·25*M* sucrose *M*/100 phosphate buffer *p*H 6·8 extract of coleoptiles grown for 48 hr. After 3 hr. dialysis against the extracting solvent, using narrow gauge dialysis tubing, the supernatant was transferred to an analytical ultracentrifuge cell and centrifuged at 260,000*g* until the 33*S* component had sedimented to the bottom of the cell. In order to clarify the serological pattern the supernatant (which should be free of the heavy components) was used to absorb out of the antiserum the antibodies specific to the relatively low molecular weight antigens, thereby producing, provided the absorption has been taken to completeness, an antiserum specific to any 'microsome' antigens. The microsome pellet was suspended in phosphate buffer

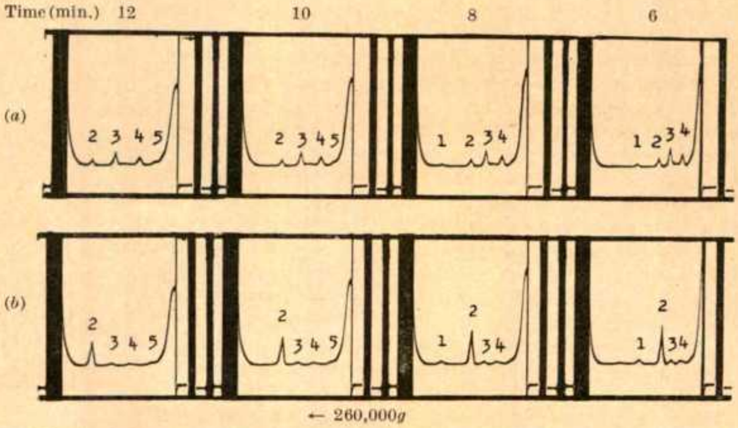

Fig. 4. Sedimentation diagrams illustrating the dissociation of the 69*S* component with *p*H change. Coleoptiles extracted in 0·25 *M* sucrose, *M*/100 phosphate buffer. *a*, *p*H 8·4; *b*, *p*H 6·8 ($S_{20,w} \times 10^{-13}$ values: peak 2 = 69; peak 3 = 50; peak 4 = 33)

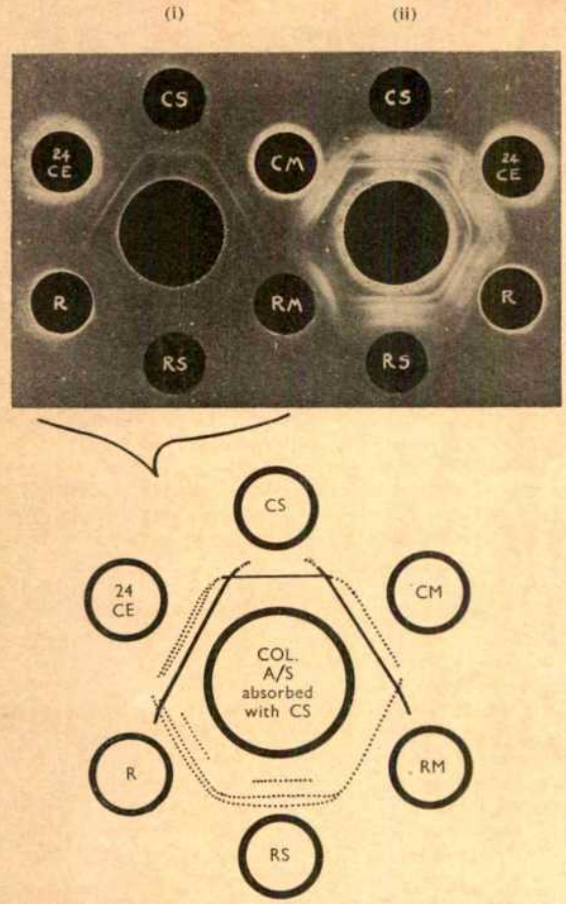

Fig. 5. Serological analyses of microsome fractions of wheat coleoptile, root and other extracts. The centre cup contained coleoptile antiserum: (i) absorbed with coleoptile extract from which the microsomes had been removed by centrifugation, and (ii) unabsorbed coleoptile antiserum. *CS*, supernatant remaining after removal of microsome particles from coleoptile protein extract by centrifugation; *CM*, coleoptile microsome fraction; *RM*, root microsome fraction; *RS*, supernatant remaining after removal of microsome particles from root protein extract; *R*, total root protein extract (that is, *RM* + *RS*); 24 *CE*, extract of coleoptiles (24 hr. after sowing) representing meristematic phase of growth

M/20, *p*H 7·6—the higher *p*H was chosen to facilitate dissociation—and the extract stored at 2° C. until required for testing.

This antiserum was afterwards tested against the microsome suspension and also a similar preparation of root microsomes. These reactions are shown with others in Fig. 5, the absorbed antiserum series on the left and the unabsorbed antiserum series on the right. To ensure that any precipitin line found to be associated with the microsome fraction was not due to incomplete absorption, a cup containing the supernatant used for absorption (*CS*) was included. The results revealed that the coleoptile microsome extract (Fig. 5i) contained two antigens when tested against the absorbed antiserum. One of these was present in the coleoptile supernatant (*CS*) and was therefore due to incomplete absorption, while the other antigen originated from the microsome fraction. In later experiments complete absorption of the antiserum was accomplished by absorbing with a larger volume of supernatant (*CS*) (Fig. 6). The position of the precipitin line associated with the microsome fraction relative to the antigen and antibody cups suggested that it was a molecule with a molecular weight of the order of thousands rather than millions. Even the monomer form of the microsome particle has a sedimentation constant in the region of 33*S*, which would imply a larger molecular weight. It would therefore appear that the 33*S* monomer must have broken down into even smaller units.

The question arises as to whether the antigenic activity resides in the typical microsome particle (69*S* + 50*S* + 33*S*) or in one or both of the 'heavy' microsome particles (peaks 0 and 1, Fig. 3*a*), or in the slower component (peak 5, Fig 3*a*). Two lines of evidence favoured the 69*S* (peak 2, Fig. 3*a*) component and its two dissociation components 50*S* (peak 3, Fig. 3*a*) and 33*S* (peak 4, Fig 3*a*) as the seat of antigenic activity; these were: (1) differential centrifugation followed by quantitative estimation[7] of the

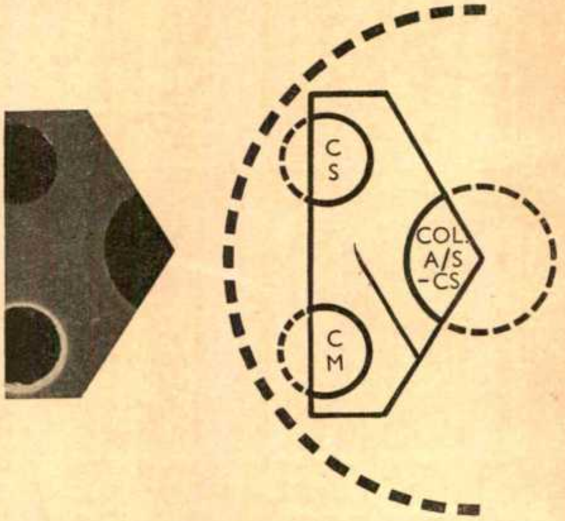

Fig. 6. Serological demonstration of an antigen associated with the microsome fraction of coleoptile protein extract. *CM*, coleoptile microsome fraction; *CS*, supernatant remaining after removal of microsome particles from coleoptile protein extract by centrifugation; *COL A/S* – *CS*, coleoptile antiserum absorbed with *CS*

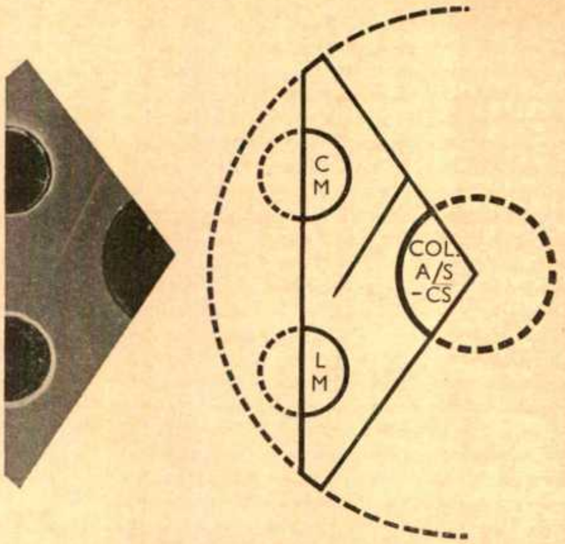

Fig. 7. Serological analyses of microsome fractions of wheat coleoptile and leaf with specific microsome fraction antiserum (that is, coleoptile antiserum (*COL. A/S*) absorbed with the supernatant (*CS*) remaining after the microsome particles have been centrifuged from a coleoptile protein extract). *CM*, coleoptile microsome fraction; *LM*, leaf microsome fraction

distribution of the antigen component; (2) the fact that the antigen still remained in the supernatant under conditions when particles of 100*S* and above (that is, including peaks 0 and 1, Fig. 3*a*) would have centrifuged down. It is quite possible that peaks 0 and 1 are higher polymers of the 69*S* particle, but in these experiments the centrifuge speed was too high to determine the $S_{20,w}$ values accurately for these two peaks. Petermann *et al.*[8] have suggested a similar hypothesis for the 'heavy' microsomes of liver extracts.

The precipitin band associated with the microsome fraction of the coleoptile extract was absent from microsome fractions of root (Fig. 5i) and leaf (Fig. 7). With the coleoptile antiserum used in these experiments any organ-specific antigens associated with root and leaf microsomes would not be detected; but it seems probable that such antigens exist.

A further point of interest was the fact that an extract made from coleoptile cells during their meristematic phase of growth (24 *CE*, Fig. 5, ii) shared nearly all its precipitin lines with the root extract (*R*, Fig. 5, ii). The exceptions were those associated with the high molecular weight components. Thus it appears, with these exceptions, that a non-organ specific meristematic pattern of antigens has superimposed upon it, during differentiation, a combination of proteins characteristic of differentiated cells.

In conclusion, it is possible to visualize the primordial cells of an organ as containing their own characteristic microsome particles as well as many antigens possessed in common with other organs. Possibly also, organ-specific microsomes synthesize organ-specific proteins or a particular combination of non-organ-specific proteins, which would either, by their incorporation into the primordial cytoplasm, or by their enzymatic activity, lead to greater cellular specialization and thereby forward the process of cellular differentiation.

I wish to express thanks to Dr. P. G. H. Gell for his many valuable suggestions throughout the course of this work, to Dr. D. R. Stanworth and Miss P. Ratcliff for carrying out the analytical ultracentrifuge determinations and to Miss A. Hudson for technical assistance demanding much patience.

These results will be reported in detail elsewhere. The work was carried out under a grant from the Nuffield Foundation

[1] Bonner, J., *Amer. J. Bot.*, **46**, 58 (1959).

[2] Ouchterlony, O., *Acta Pathol. Microbiol. Scand.*, **25**, 186 (1948).

[3] Brown, R., and Rickless, P., *Proc. Roy. Soc.*, B, **136**, 110 (1949).

[4] Freund, J., and Bonanto, M. V., *J. Immunol.*, **48**, 325 (1944).

[5] Ts'o, P. O. P., Bonner, J., and Vinograd, J., *J. Biophys. Biochem. Cytol.*, **2**, 451 (1956).

[6] Ts'o, P. O. P., Bonner, J., and Vinograd, J., *Biochim. Biophys. Acta*, **30**, 570 (1958).

[7] Wright, S. T. C., *Nature*, **183**, 1282 (1959).

[8] Petermann, M. L., Mizen, N. A., and Hamilton, M. G., *Cancer Res.*, **16**, 620 (1956).

SPECTROPHOTOMETRIC MEASUREMENT OF CYTOCHROMES IN THE RED AND THE WHITE MUSCLES OF THE AMERICAN COCKROACH, *PERIPLANETA AMERICANA* L.

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SINCE the early report by Sacktor and Bodenstein[1], the red muscle of the male and the white one of the female American cockroach, *Periplaneta americana* L., have long been a subject of investigation, and parallelism between the coloration and the activity of oxidative enzymes of the muscles has been reported[2]. However, ordinary spectrophotometric measurement of the coloration was precluded by the turbidity of muscle. In the present experiment, the opal glass transmission method of Shibata[3] was adopted, instead of the usual spectroscopic method, which is subject to rather large errors, in an attempt to obtain quantitative or at least semi-quantitative data on the suspected difference in the content of cytochromes between the red and the white muscles of the American cockroach.

For observation of the muscle, red thoracic trochantinal and coxal muscles were collected from meso- and meta-thoraces of male adult insects; only the 'pink' muscles[4], which are identical with the red muscles of the male, were collected from the female thoraces. The muscles were soaked in 0·01 *M* diethyldithiocarbamate solution to prevent formation of melanin, which might occur. 30 mgm. of the solution was used for 100 mgm. of fresh muscle. The sample was kept in a Petri dish with a sheet of wet filter paper to avoid drying during the dissection. Then the sample was lightly ground with a small glass rod and a small amount of hydrosulphite was added.

For observation of the mitochrondria, muscles collected as above were homogenized in a glass homogenizer as a 2 per cent suspension in a solution of *p*H 7·4, containing 0·25 *M* sucrose, 0·05 *M* phosphate buffer and 0·01 *M* sodium ethylenediaminetetraacetate. On centrifuging the homogenate at 600*g* for 5 min., the precipitate was homogenized in a smaller quantity of the solution and the process was repeated five times. At the fifth extraction very few mitochondria were obtained. The resultant supernatant solution was centrifuged at 10,000*g* for 10 min. and the precipitate was washed twice by re-suspending and re-centrifuging. The procedures described above were carried out at about 4° C. The final suspension was shown microscopically to contain almost pure mitochondria, which were stained with janus green *B*. Mitochondria thus obtained show oxidative activity with α-glycerophosphate, succinate, L-glutamate, fumarate, L-malate, α-keto-glutarate and oxalacetate as substrates (un-

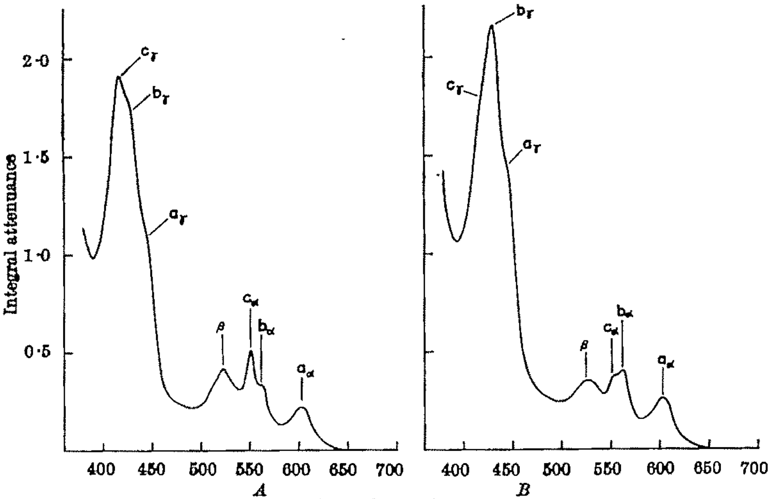

Fig. 1. Integral attenuance spectrum of (*A*) reduced red muscle of male adult American cockroach and (*B*) reduced mitochondria extracted from red muscle of male adult American cockroach, showing absorption maxima of $\alpha(a_\alpha, b_\alpha, c_\alpha)$, $\beta(\beta)$ and Soret $(a_\gamma, b_\gamma, c_\gamma)$ bands of cytochromes

published results). The final precipitate was left to drain for 1 min. with the centrifuging tube upside down. A few crystals of sodium hydrosulphite were added to the precipitate and thoroughly mixed with it.

Although muscle and mitochondria diffused light considerably to give clear absorption spectra, opal glass was used to make sure. A brass plate, 0·6 mm. thick, with a hole a little larger than the section of the incident beam of the spectrophotometer, was placed on a plate of opal glass. The sample was put in the hole of the brass plate and a plate of transparent glass was pressed on to the sample to form an optical cell containing the sample of a definite thickness. This was mounted on an opal glass attachment and the spectrum was taken in the region 380–700 mμ. From 380 to 500 mμ black tape was applied on the reference side to lower the base-line height of the spectrum.

Since spectra of semi-integral attenuance thus obtained were lowest and constant beyond 660 mμ, this constant base-line height was subtracted to obtain integral attenuance[3], assuming that the ratio of reflectance to transmittance was approximately constant in the wave-length range observed. The spectra showed the presence of cytochromes *a*, *b* and *c* both in the muscle and in the mitochondria. Typical spectra of the muscle and the mitochondria of the male cockroach are shown in Fig. 1. Spectra of the female preparations were qualitatively much the same as those of the male.

Table 1. INTEGRAL ATTENUANCE OF REDUCED MUSCLE AND MITOCHONDRIA AT ABSORPTION MAXIMA OF THE α-BANDS OF CYTOCHROMES

	Absorption maximum (mμ)	Integral attenuance		Male/Female
		Male	Female	
Muscle				
Cytochrome *a*	603	0·278	0·065	4·28
Cytochrome *b*	562	0·422	0·122	3·47
Cytochrome *c*	551	0·587	0·164	3·58
Mitochondria				
Cytochrome *a*	603	0·259	0·181	1·43
Cytochrome *b*	562	0·399	0·305	1·31
Cytochrome *c**	551	0·357	0·297	1·20
Dry matter of mitochondria (per cent)		19·5	18·6	

* Values for cytochrome *c* were reduced by washings

Though the absorption maximum of cytochrome *b* at about 562 mμ was obscured by the distinct absorption band of cytochrome *c*, which has its maximum at about 551 mμ, the attenuance at 562 mμ is considered to represent roughly the amount of cytochrome *b*, since cytochrome *c* absorbs light very little at this wave-length. For example, absorption at 562 mμ is about 10 per cent of that at 550 mμ in the case of the cytochrome *c* extracted from the thoracic muscle of the horn beetle, *Xylotrupes dichotomus* L. (unpublished results). In the case of mitochondria, however, cytochrome *c* was considerably washed out and the peaks of cytochrome *b* became rather clear both in the α region and in the Soret region; and cytochrome *b* was seen to remain in the mitochondria as well as cytochrome *a*. Experimental results are given in Table 1.

In all cases with muscle, the attenuance ratio of male to female was higher for cytochrome *a* than for cytochrome *c* or *b*. Though the values vary somewhat, probably with age of cockroaches, it is apparent that red muscle contains a much greater quantity of cytochromes than white muscle. On the other hand, there was no great difference in the concentration of cytochromes in mitochondria between male and female, both on a wet- and a dry-weight basis. As for the nature of mitochondria, it is not yet determined whether the mitochondrial fraction sedimented by the procedure employed is the only fraction that contains cytochromes, especially *a* and *b*. However, it seems likely that the difference in the content of cytochromes between the red and the white muscles is largely due to a greater amount of mitochondria in red muscle than in white muscle, and not so much to the difference in the concentration of cytochromes in mitochondria. In fact, red muscle yielded more than three times as much mitochondria as white muscle, both on a wet- and a dry-weight basis.

Though further experiment must be carried out to determine what substances contribute to the reddish coloration of the muscle of the American cockroach, I believe that cytochromes *a*, *b* and *c* are largely responsible. So far, I have not been able to detect any other substance in the intact and reduced muscles from their spectra.

Experiments are now in progress by this method to compare the content of cytochromes in muscles of various species of insects, and the results will be published elsewhere.

I thank Dr. J. Fukami, of this Laboratory, Mr. S Matsunaka of the Section of Physiology and Genetics, for advice, and Dr. H. Ishikura, chief of this Laboratory, for his interest and encouragement.

[1] Sacktor, B., and Bodenstein, D., *J. Cell. Comp. Physiol.*, **40**, 152 (1952).

[2] Fukami, J., *Oyo-Kontyu*, **12**, 8 (1956).

[3] Shibata, K., "Methods of Biochemical Analysis", **7**, 77 (1959)

[4] Brooks, M. A., *Ann. Entom. Soc. Amer.*, **50**, 122 (1957).

LETTERS TO THE EDITORS

COSMOLOGY

Pasteur and Cosmic Asymmetry

Yang and Lee's[1] discovery of the non-conservation of parity in certain interactions of "fundamental particles" has led to a general acceptance of the notion that the structure of the universe is asymmetrical. So far as I know this was first enunciated by Pasteur[2]. The following are his exact words :

"Il faut de toute nécessité que des actions dissymétriques président pendant la vie à l'élaboration des vrais principes immédiats naturels dissymétriques.

"Quelle peut être la nature de ces actions dissymétriques ? Je pense, quant à moi, qu'elles sont d'ordre cosmique. L'univers est un ensemble dissymétrique et je suis persuadé que la vie, telle qu'elle se manifeste à nous, est fonction de la dissymétrie de l'univers ou des conséquences qu'elle entraîne. *L'univers est dissymétrique.*"

The first sentence is, I think, the first enunciation of the principle that an asymmetrical phenomenon must have an asymmetrical cause. It would be of great interest to know why Pasteur, who believed that there was a sharp gap between vital and non-living processes, attributed the asymmetry to the universe, and not to a 'vital force' or some such agency acting in parts of it. Pasteur's conjecture may of course be disproved in either or both of two ways, namely by the discovery on other planets of living beings composed mainly of sugars and amino-acids of the D series, or by the synthesis of such beings by men. But until such disproof is forthcoming he deserves full credit for his formulation.

I have to thank Prof. Jacques Monod, of the Institut Pasteur, for most kindly furnishing me with Pasteur's exact wording, and for a valuable comment.

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Nov. 18.

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[1] Yang, C. N., and Lee, T. D., *Phys. Rev.*, 104, 257 (1956).

[2] Pasteur, L., *C.R. Acad. Sci., Paris* (June 1, 1874) (reprinted in "Œuvres", 1, 361).

GEOPHYSICS

Effect of Latitude on the Diurnal Maximum of 'Dawn Chorus'

IT is known that chorus, or 'dawn chorus', exhibits a pronounced diurnal variation in its occurrence and that different stations show different local times for the diurnal maxima. These maxima have been shown to be a function of geomagnetic latitude by Allcock[1] and independently by Pope[2].

The observations used were samples taken at 3-hr. intervals, with a consequent limitation in time resolution (excepting those obtained by the College station). The availability of observations from a large number of stations with a time resolution of 1 hr. during the International Geophysical Year makes it desirable to reconsider the latitude effect.

The data were obtained in various ways. The Geophysical Institute operated field-stations at College and Kotzebue. The data for Dunedin, Wellington, Stanford, Boulder, Seattle, Anchorage and part of those from Unalaska were supplied by IGY Whistlers-West. The data from Bermuda, Washington, D.C., Norwich, Mont Joli, Knob Lake and part of those for Unalaska were supplied by IGY Whistlers-East. The Cambridge point was obtained from Storey's doctoral dissertation[3]. The time of maximum at Saskatoon was supplied by G. M. Allcock, of New Zealand. The Macquarie result was communicated directly by radio from that station.

With the exception of the last three stations, which used the occurrence of chorus, the diurnal curves were determined from hourly chorus indices. These indices are derived through a subjective estimate of the strength of the phenomenon. Using the strengths can be considered a weighting of the occurrences in favour of those close to the station. The difference between these two techniques of obtaining the diurnal maximum is probably small.

Table 1 gives the various 'whistler' stations used in this work along with the parameters involved.

Fig. 1 is a plot relating the local time of the diurnal maximum to geomagnetic latitude. The arrows attached to the data points indicate the extent and direction of correction for magnetic time. Observations from the eastern stations are plotted with solid points while those from the western stations are plotted as open points. It is apparent that the eastern stations tend to the left of the plot whereas the western tend to the right side. Thus there appears to be an 'east–west effect' in the time of diurnal maximum, which is probably a spurious effect.

Maeda[4] has considered the effect of a distortion of the Earth's magnetic field by rotation to explain the westward shift of the cosmic-ray equator noted by Simpson[5]. This westward shift would tend to reduce the east–west effect in the northern hemisphere. The Knob Lake station, being closer to the magnetic equator, would have a lower latitude, while the

Table 1

Abbreviation	Station	Geomagnetic latitude	Eccentric geomagnetic latitude	Local time maximum	No. of months
CO	College, Alaska	64·9	67 5	14 0	36
KO	Kotzebue, Alaska	63·9	67·1	13·2	13
KL	Knob Lake, Canada	66·1	63·2	8·5	4
UN	Unalaska, Alaska	51·1	54·2	6·4	9
SE	Seattle, Washington	53·8	54 5	6·8	10
ST	Stanford, California	43·8	45·7	3·9	7
BO	Boulder, Colorado	48 9	48 3	5 5	8
NR	Norwich, Vermont	55·1	52·5	6 0	14
MJ	Mont Joli, Canada	60·0	57·1	7·5	9
WA	Washington, D.C.	50·2	47·8	3·4	10
BE	Bermuda	43·7	40·7	2·7	7
CA	Cambridge, England	54·7	52·1	6·0	3
SA	Saskatoon, Canada	60·5	60·3	9·6	1
MQ	Macquarie Island	60·2	64 4	11·0	—
DU	Dunedin, N.Z.	50·5	54·3	4·9	3
WE	Wellington, N.Z.	45·2	48·9	4 2	5

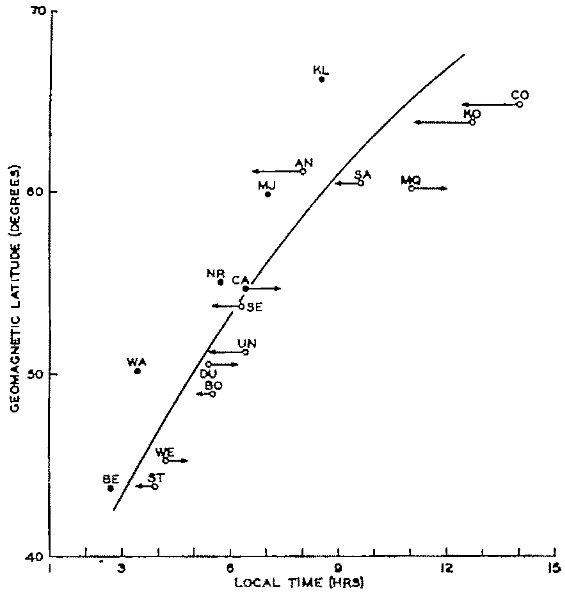

Fig. 1. Time of diurnal maximum versus geomagnetic latitude

College station, being farther from the equator, would have a higher latitude. This qualitative explanation, however, is not successful if the three southern hemisphere stations are considered, since they are all increased in latitude. Therefore, accepting the validity of the observations from the southern hemisphere stations, this westward shift of the geomagnetic equator fails to resolve the discrepancy without introducing others.

The use of geomagnetic latitudes derived from dip angles also fails to remove the discrepancy.

More success can be had by use of an eccentric dipole field. Parkinson and Cleary[6] have computed the position of the eccentric dipole-field poles for the epoch 1955. They are 81·0° N., 84·7° W. and 75·0° S., 120·4° E. Since these poles are not antipodal, the problem of computing effective geomagnetic latitudes is complicated. A simple approximation can be obtained, however, by computing the latitudes in each hemisphere separately as if the dipole were centred, using the appropriate pole. This approximation will, of course, produce a discontinuity at the equator. The latitudes using this approximation are tabulated and the results plotted in Fig. 2. The magnetic time corrections are plotted in the same manner as in Fig. 1. It is clear that while some scatter remains, the east–west effect has disappeared. In particular, the Knob Lake point is brought into reasonable agreement with the rest of the observations.

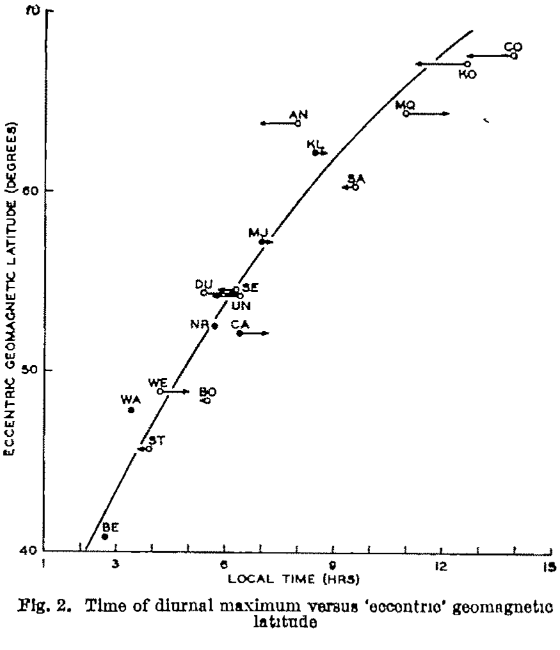

Fig. 2. Time of diurnal maximum versus 'eccentric' geomagnetic latitude

These results indicate that it is desirable to consider the use of the eccentric dipole field in connexion with theories of chorus. Also, the paths of whistler propagation may be affected by the eccentric field rather than the centred field, which may be an important consideration from the point of view of conjugate point experiments and electron density computations.

This work was supported by the U.S. Air Force Cambridge Research Center under Contract No. *AF* 19(604)–5574. The observations from College and Kotzebue during the International Geophysical Year were obtained under a National Science Foundation Grant (*IGY* 6.10). The comments of Dr. L. Owren are gratefully acknowledged.

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[1] Allcock, G. M., *Austral. J. Phys.*, **10**, 286 (1957).

[2] Pope, J. H., *Nature*, **180**, 433 (1957).

[3] Storey, L. R. O., Ph.D. dissertation, University of Cambridge.

[4] Maeda, Ken-Ichi, *Ionospheric Research in Japan*, **11**, 116 (1957).

[5] Simpson, J. A., Geophysical Monograph No. 1 (1956).

[6] Parkinson, W. D., and Cleary, J., *Geophys. J.*, **1**, 346 (1958).

Calculation of Cosmic-ray Ages in the Iron Meteorites 'Carbo' and 'Treysa'

In a recent communication[1], Dr. R. R. Marshall estimates the cosmic-ray age of the iron meteorite 'Carbo' to be 0·69 × 10^9 yr. His calculation was based upon the following experimental results of ours[2] and Barr's[3]. We showed that the relative isotopic abundances of potassium from a sample of the iron meteorite 'Carbo' were markedly different from those of terrestrial potassium. Considerable enrichment of the isotopes of masses 40 and 41 was found ($^{39}K_m$ = 79·5 per cent, $^{40}K_m$ = 4·8 per cent, and $^{41}K_m$ = 15·7 per cent as compared to 93·08, 0·012 and 6·91 per cent respectively for ordinary terrestrial potassium). Evidence was given that a considerable amount of this meteoritic potassium—nearly all the potassium-40 and more than half the potassium-41—is of cosmogenic origin, that is, it is a reaction product of the spallation of mainly iron nuclides, induced by high-energy particles from cosmic radiation. Barr[3] measured the cross-sections for the production of fifty-nine radioactive isotopes formed by 5·7-BeV. proton bombardment of copper. From his summary of measured and interpolated independent cross-sections, Marshall calculated the total cross-sections for the production of the potassium isotopes to be σ_{39} = 14·8 mb., σ_{40} = 9·4 mb., and σ_{41} = 14·7 mb.

The method of determining the cosmic-radiation age of meteorites is based on the fact that a stable and a radioactive isotope (potassium-41 and potassium-40) are formed at the same time. During the time the meteorite is exposed to cosmic radiation, the ratio of potassium-41 to potassium-40 increases with time due to the radioactive decay of potassium-40. Besides the difficulty of measurement of isotopic abundance on the small amounts of meteoritic potassium available, the exact determination of such a potassium-41/potassium-40 age for a given meteorite is mainly a question of knowing the ratio σ_{41}/σ_{40} of the production cross-section for potassium by cosmic-ray bombardment of iron meteorites. The use of observations from a 5·7-BeV. proton bombardment of copper for the age determination of iron meteorites can only be considered as an approximate solution of the problem.

In a re-calculation of the cosmic-radiation age from the preceding values for the relative isotopic abundances and the production cross-sections, and with a value of λ taken as $0{\cdot}54 \times 10^{-9}$ yr.$^{-1}$, we found the cosmic-ray age of 'Carbo' to be $T = 1{\cdot}36 \times 10^9$ yr., instead of $0{\cdot}69 \times 10^9$ yr. as calculated by Marshall. We both used the same formula and practically the same numerical constants, with the exception that we defined R as the ratio potassium-39/potassium-41 in ordinary potassium instead of potassium-39/potassium-40 as is defined in Marshall's paper. As this difference alone cannot explain the discrepancy in the two results of age calculation, there appears to be another error in Marshall's work which we are not quite able to understand.

As the results of our isotopic abundance measurements on the extremely small potassium samples available for measurements still show rather large statistical fluctuations[4], the uncertainties in the cosmic-radiation ages are relatively large. We calculate the radiation age T from the equation (see equation (3) in our paper[4]):

$$\frac{\lambda T}{1-\exp.(-\lambda T)} = \frac{({}^{41}\mathrm{K}_m - a\,{}^{39}\mathrm{K}_m)/{}^{40}\mathrm{K}_m}{\dfrac{\sigma_{41}}{\sigma_{40}}\left(1 - a\,\dfrac{\sigma_{39}}{\sigma_{41}}\right)} \tag{1}$$

where a is ${}^{41}\mathrm{K}/{}^{39}\mathrm{K}$ in ordinary potassium (this corresponds to Marshall's equation if $R = 1/a = {}^{39}\mathrm{K}/{}^{41}\mathrm{K}$ and not ${}^{39}\mathrm{K}/{}^{40}\mathrm{K}$). From the scattering in the experimental results for $({}^{44}\mathrm{K}_m - a\,{}^{39}\mathrm{K}_m)/{}^{40}\mathrm{K}_m$ (1·90 ... 2·05 for potassium from 'Carbo' and 1·65 ... 1·72 for potassium from 'Treysa'), we obtain the following limits for the radiation ages of the iron meteorites 'Carbo' and 'Treysa', using the cross-sections as given above:

$$\begin{aligned} &\text{'Carbo'}: \quad T_C = (1{\cdot}05 \ldots 1{\cdot}40) \times 10^9 \text{ yr.} \\ &\text{'Treysa'}: \quad T_T = (0{\cdot}50 \ldots 0{\cdot}67) \times 10^9 \text{ yr.} \end{aligned} \tag{2}$$

The uncertainty in the assumptions upon which the age determination is based and particularly the adoption of a relatively uncertain value for the ratio of σ_{41}/σ_{40} still require a very reserved interpretation of these ages. This is the reason why, in our own treatment of the potassium-41/potassium-40 age problem[4], we eliminated the production cross-sections. In that way we obtained the following relation for the difference of the cosmic-ray age of 'Carbo' and 'Treysa' only:

$$0{\cdot}35 \times 10^9 \text{ yr.} < T_C - T_T < 1{\cdot}25 \times 10^9 \text{ yr.} \tag{3}$$

The rather wide range for this difference depends upon the scattering of experimental results and on the possibility that the absolute radiation age of 'Carbo' may have any value between several times 10^8 and $4{\cdot}5 \times 10^9$ yr. so long as no special assumptions are made regarding the production cross-sections. The value of the upper limit of the difference holds if the radiation age of 'Carbo' should be as high as $4{\cdot}5 \times 10^9$ yr., as is theoretically possible (although very unlikely). The estimate (3) is consistent with the preceding results (2) on the radiation ages themselves, but has a much higher degree of certainty since the statement is completely independent of the ratio σ_{41}/σ_{40}. The existence of such a difference of several hundred millions of years for the radiation ages of iron meteorites is in itself a remarkable fact.

We wish to thank Dr. F. Begemann of this Institute for valuable discussions.

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[1] Marshall, R. R., *Nature*, **184**, 117 (1959).

[2] Voshage, H., and Hintenberger, H., *Z. Naturforsch.*, **14a**, 194 (1959).

[3] Barr, D. W., thesis, University of California, Radiation Laboratory Report 3793 (1957).

[4] Voshage, H., and Hintenberger, H., *Z. Naturforsch.*, **14a**, 828 (1959).

IN the last sentence of the second paragraph of my original, ${}^{40}\mathrm{K}$ was misread for ${}^{41}\mathrm{K}$; the definition should read: R is the ratio of ${}^{39}\mathrm{K}/{}^{41}\mathrm{K}$. Similarly, at the end of par. 4, σ_{29} should read σ_{39}, and in par. 5, line 17, σ_{40} should read σ_{39}. An error in the numerical evaluation of the radiation age led to a figure exactly half that of the correct value. Therefore, in both par. 5, line 2, and in par. 7, line 6, $0{\cdot}6_9$ should read $1{\cdot}3_8 \times 10^9$ years. Fortunately, these corrections do not affect either the principle of the calculation or, in general, the comparisons drawn in this letter.

I am grateful to Dr. H. Voshage and Dr. H. Hintenberger for directing attention to these discrepancies.

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RADIOPHYSICS

Solar Radio Bursts and Low-energy Cosmic-rays

THE recent discovery, by equipment carried in satellites and balloons, that the terrestrial atmosphere is frequently bombarded by solar protons of energies 30–300 MeV.[1,2], has given rise to much speculation concerning the origin of these low-energy cosmic rays, and their relation to solar radio bursts, flares, and terrestrial disturbances. In this communication we seek to elucidate these problems from a consideration of radio spectral observations of the Sun. The radio data were obtained at the Harvard Radio Astronomy Station, Fort Davis, Texas, with sweep-frequency receivers[3] covering the range 25–580 Mc./s. The equipment scans the entire frequency-range three times per second, and a continuous record of solar radio bursts has been obtained from sunrise to sunset for the past three years.

The observations show that the radio bursts which are associated with the ejection of low-energy cosmic rays from the Sun take the form of an intense continuum, sometimes known as spectral Type IV. This radiation characteristically covers several octaves of the radio spectrum, shows only gradual changes in intensity and frequency-range, and often lasts for many hours. It probably originates in a synchrotron mechanism[4], in which electrons with relativistic velocities are trapped in orbital motion in a magnetic field. About 15 such bursts have been recorded at Fort Davis each year during the solar maximum.

The continuum radiation is related to the ejection of low-energy cosmic rays from the Sun by a sequence of events which commences with a solar flare, usually of class 3 or 3+ (ref. 5). The flare is followed, with an average delay of 12 min., by an outburst of continuum radiation, and approximately 1 hr. later an enhanced flux of protons arrives at the Earth. Their energies, as measured by Geiger and scintillation counters carried in balloons and satellites, lie in the range 30–300 MeV., which corresponds to particle velocities of 0·2–0·7 c. Because of their low rigidity, the protons can penetrate the terrestrial atmosphere only in the vicinity of the magnetic poles, where they give rise to ionospheric absorption of very-high-frequency radio waves over the polar cap[6,7]. These polar blackouts, resulting from enhanced ionization in the *D* region of the ionosphere, at present constitute the best parameter for measuring the arrival time of the solar particles at the Earth. The minimum time delay between the onset of the solar continuum radiation and the polar blackouts is 0·7 hr.

It may be suggested that at the time the optical flare is observed, a stream of protons and electrons is ejected violently outwards from the flare region, at a velocity of approximately 0·5 c. The electrons in this stream are afterwards trapped in orbital motion in a magnetic field in the coronal levels above the active area, and generate energy in the radio band of electromagnetic spectrum. (The strength of the magnetic field, as indicated by the splitting of slow-drift radio bursts, is of the order 1–10 gauss.) The protons are not captured by the magnetic field and continue to travel outwards at high velocity, spiralling along elongated lines of force that stream from the Sun into the interplanetary plasma[8]. A travel time of 0·7 hr. over a direct path between the Sun and the Earth corresponds to a particle velocity of 0·2 c. For spiral paths the velocities will be proportionately higher. Such velocities are in accord with those deduced from direct measurements of the particle energies.

The heliographic distribution of the flares associated with continuum radiation is given in Fig. 1, and their importance distribution in Table 1. It will be seen that there is a highly significant western excess of flares associated with the ejection of low-energy cosmic rays that afterwards reached the Earth, and that most of these flares were of importance 3. The western excess has also been noted by Reid and Leinbach[7], who surmise that the connective magnetic lines of force between the Sun and the Earth emanate mainly from the western hemisphere of the Sun. These are possibly the remnants of earlier auroral corpuscular streams which have followed the well-known curved path described in the early literature by Chapman[9].

The solar protons have now been detected directly by satellite and balloon equipment on nine separate occasions. In all cases they were associated with ionospheric absorption phenomena in the polar regions. The available radio data also show that the emission of these solar protons is always preceded by outbursts of continuum radiation. In approximately 75 per cent of cases they have also been succeeded, within a few days, by a geomagnetic storm, and a Forbush decrease in extra-solar cosmic-ray intensity. There is at present some disagreement concerning the relation of magnetic storms to solar radio bursts, but a detailed analysis[10] of the phenomena suggests that the emission of auroral streams, that is, particles with velocities of 1,000 km./s. (0·003 c.), is not primarily associated with solar continuum radiation, but rather with the occurrence of slow drift (Type II) radio bursts. Further analysis also suggests that noise storm bursts (Type I) and fast drift bursts (Type III) are inconsequential with regard to the terrestrial disturbances considered in this communication.

Table 1. Importance Classification of Flares associated with Continuum (Type IV) Radio Bursts, May 1, 1957–July 31, 1959

Importance of associated flare	Number of continuum outbursts	
	Followed by polar blackouts	Not followed by polar blackouts
3, 3+	8	3
2, 2+	2	9
1, 1+	0	7

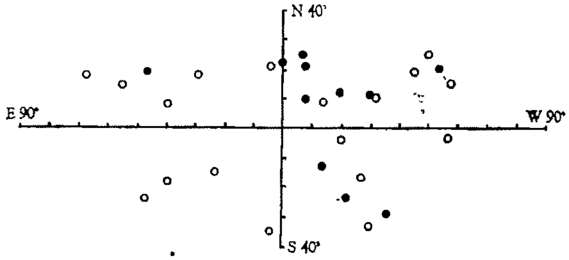

Fig. 1. Heliographic distribution of flares associated with continuum (Type IV) radio bursts, May 1, 1957–July 31, 1959. ●, Followed by ionospheric polar blackout; ○, not followed by ionospheric polar blackout. The polar blackout data are taken from Reid and Leinbach (ref. 7)

This work forms part of a research programme supported by financial assistance from the Geophysics Research Directorate of the United States Air Force, through the offices of Sacramento Peak Observatory.

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[1] Rothwell, P., and McIlwain, C., *Nature*, 184, 138 (1959).

[2] Anderson, K. A., Arnoldy, R., Hoffman, R., Peterson, L., and Winckler, J. R., *J. Geophys. Res.*, 64, 1133 (1959).

[3] Maxwell, A., Swarup, G., and Thompson, A. R., *Proc. Inst. Radio Eng.*, 46, 142 (1958).

[4] Boischot, A., and Denisse, J. F., *C.R. Acad. Sci., Paris*, 245, 2194 (1957).

[5] Thompson, A. R., and Maxwell, A., *Planet. Space Sci.* (in the press).

[6] Reid, G. C., and Collins, C., *J. Atmos. Terr. Phys.*, 14, 63 (1959).

[7] Reid, G. C., and Leinbach, H., *J. Geophys. Res.*, 64, 1801 (1959).

[8] Gold, T., *Nature*, 183, 355 (1959).

[9] Chapman, S., *Mon. Not. Roy. Astro. Soc.*, 89, 456 (1929).

[10] Maxwell, A., Thompson, A. R., and Garmire, G., *Planet. Space Sci.* (in the press).

PHYSICS

Variation with Temperature of the Magnetic Susceptibility of Some Transition Elements

The dependence on temperature of the magnetic susceptibilities of the transition elements has been measured over various ranges of temperatures[1], and a series of experiments is now being carried out in this Department in an attempt to confirm some of the results and extend others in the temperature range 20°–1,800° C. Preliminary results have been obtained for the elements platinum, rhodium,

Table 1

Temp. (° C.)	Susceptibility (e.m.u. gm.$^{-1}$ × 10^{-6}) Ti	Zr	Nb	Ta	Rh	Pt
20	3·35	1·32	2·28	0·85	1·05	0·97
250	3·65	1·36	2·08	0·83	1·13	0·85
500	4·18	1·39	2·00	0·80	1·25	0·73
750	4·26	1·45	1·90	0·77	1·37	0·64
1,000	5·39	1·64	1·81	0·75	1·48	0·56
1,250	5·25		1·74	0·73	1·56	0·50
1,500			1·71	0·72	1·64	0·45
1,750			1·69	0·715		0·40

zirconium, titanium, tantalum and niobium. The room temperature values of the susceptibilities (measured by comparison with platinum at room temperatures) and the values at temperature intervals up to 1,750° C. are given in Table 1.

The elements used were Johnson–Matthey 'Specpure', and no appreciable dependence of susceptibility on field-strength was observed. The error in the room temperature values is estimated as $\pm 0{\cdot}05 \times 10^{-6}$ e.m.u. gm.$^{-1}$. The values given in Table 1 for titanium and zirconium are in quite good agreement with the results of Squire and Kauffmann[2] (the temperatures at which the discontinuities in the $\chi - T$ curves occur being 860° C. for titanium and 880° for zirconium). The values for niobium and tantalum are similar to those of Kriessman[3].

These results supplement the evidence cited by Kriessman and Callen[1] to show that the dependence on temperature of the susceptibility of the transition elements has the same general form for each element in a particular column of the Periodic Table.

Measurements are being made on other transition elements, and a fuller account will be submitted elsewhere.

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[1] Kriessman, C. J., and Callen, H. B., *Phys. Rev.*, **94**, 837 (1954).

[2] Squire, C. F., and Kauffmann, A. R., *J. Chem. Phys.*, **9**, 673 (1941).

[3] Kriessman, C. J., *Rev. Mod. Phys.*, **25**, 122 (1953).

Surface Characteristics of Fractured Poly(methyl methacrylate)

A study of fracture phenomena in poly(methyl methacrylate) has been undertaken, which involves the breaking, in tension, of strip samples, each containing a preformed crack which lies in a plane normal to the long axis (Fig. 1). The samples are extended at fixed rates of elongation until rupture ensues, invariably at the site of the initial crack. It appears by visual inspection that the fracture process occurs in two distinct stages. The crack starts to grow fairly slowly at a velocity which is of the order of 0·1 cm./sec., until a critical stage is reached when the velocity suddenly increases to a value of at least 10^4 cm./sec. It is not yet known what factor is critical in producing the transition. The final high velocity is possibly the theoretical maximum, limited by the velocity of a longitudinal wave in the medium[1], but no accurate measurements have yet been made. A sharp characteristic audible effect is associated with the rapid stage of crack growth.

On examination of the fracture surface, the regions corresponding to the above behaviour can be readily distinguished. Two matching fracture surfaces (that is, from the same sample) are shown in Fig. 2. In the region of slow crack growth, the surface is irregular, with deep lines of level differences, usually running in the direction of crack propagation. The increase in crack velocity is indicated by a change in the character of the surface. It becomes smoother and highly reflecting, with only shallow lines of level-differences. In this 'fast' region the degree of roughness increases, and the occurrence of the familiar parabolic figures[2] obtrudes as the distance from the initial crack increases, though this transition is gradual. The area occupied by these separate regions decreases as the length of the initial crack decreases; and in samples which do not contain deliberate cracks initially, the regions lie so close to the origin of the crack that they cannot readily be distinguished.

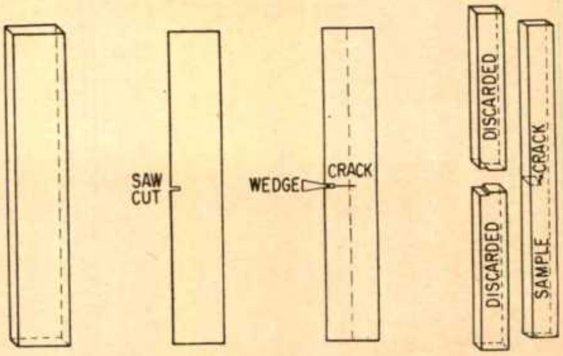

Fig. 1. Method of preparation of tensile samples (the position of the broken line in the third stage, along which the strip is cut to give the sample, determines the depth of the crack)

Perhaps the most surprising feature of the fracture surfaces formed under these conditions is the colour effects which are observed. These are readily visible to the naked eye, but are best seen under a low-power microscope (× 10) with vertical illumination. In the highly reflecting region, fringes or bands are observed, the colours of which vary in regular order with increasing distance from the initial crack. Second- and possibly higher-order 'spectra' can usually be seen before the effect is lost because of increasing roughness of the surface. Careful examination of the fringes on the fracture surface of one-half of the broken sample reveals that they do not run continuously across the surface, but occupy alternate regions bounded by the lines of level-differences.

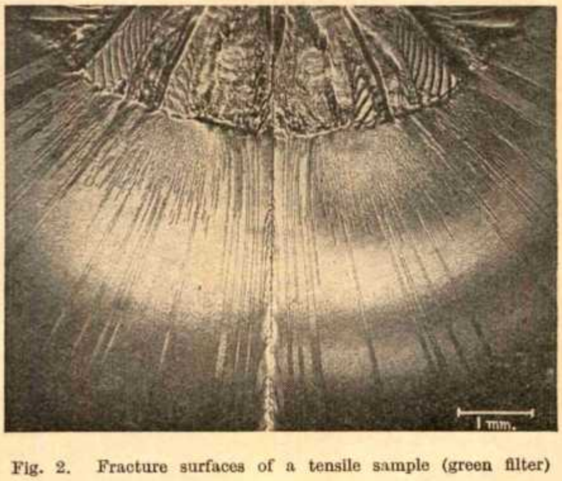

Fig. 2. Fracture surfaces of a tensile sample (green filter)

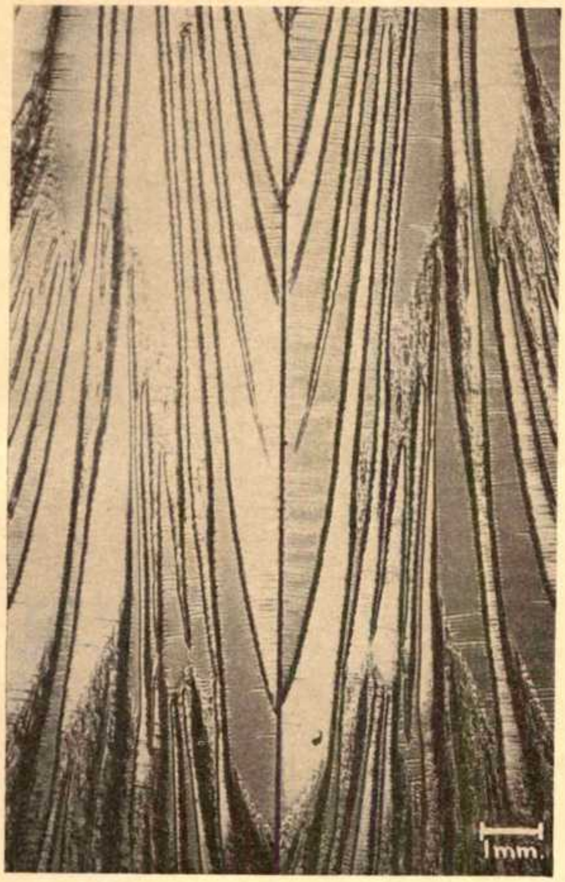

Fig. 3. Fracture surfaces of a cleavage sample (green filter)

In the intervening regions, a second set of fringes can be observed, the colours of which appear to be complementary to the first set. If the two matching fracture surfaces are examined simultaneously (Fig. 2), this duality persists in the second half of the sample, but the order of the fringes is inverted. Thus the colour fringes in the corresponding areas on the two surfaces, as defined by the lines of level-differences, exhibit the same complementarity found in adjacent regions on any one of the surfaces.

In the initial region, where the crack is growing slowly, colour effects are also observed, but they can best be produced under conditions where the rate of growth of the crack can be more readily controlled, as in a cleavage experiment (Fig. 3). In this case, fringes are not produced, and the colours are remarkably constant in hue over large areas. The same phenomenon of the complementary colours in corresponding and adjacent regions is still present, however. Colour effects have also been produced by an impact type of fracture process, as recently reported by Wolock[3].

It is believed that some of the observations noted above can be ascribed to optical interference at the surface. As the crack progresses through the (glassy) polymer, the high stresses at the tip will tend to orient the macromolecules in a direction parallel to the applied stress and hence normal to the direction of the crack and the final fracture surface. The extent to which orientation occurs will depend on the magnitude of the stress at the tip of the crack, the time for which it is applied and hence the instantaneous velocity of the crack, the temperature at the crack tip, and the viscous properties of the polymer. The refractive index along the individual polymer chains is different from that in a perpendicular direction. Hence, if the orientation of the polymer chains extends down to a specific depth into the bulk material, the result will be the same as that of a thin film on a substrate of different refractive index. This can therefore give rise to interference colours. The occurrence of colour fringes in the region of rapid crack growth can be interpreted as changes in the thickness of the film, that is, extent of orientation, as the crack progresses. In the region where the crack is growing slowly (1 mm./min. in a cleavage experiment), the film would be considered to be of remarkably constant thickness.

The orientation hypothesis is supported by other evidence. Thus the colours are thermolabile, and are destroyed by solvent in the liquid or vapour phase. There is a great discrepancy between the observed ($\sim 4 \times 10^5$ ergs/cm.^{2})[4] and calculated ($\sim 10^3$ ergs/cm.^{2}) surface energy for this material, the latter value having been computed from the energy required to rupture the chemical bonds in unit area of cross-section, assuming perfect orientation (cf. ref. 5). Clearly, a considerable amount of energy is required to produce the extent of orientation necessary to give the interference effect postulated above. However, evidence for the orientation hypothesis is still far from complete, and no reasonable explanation has been formulated for the complementarity effect which has been described above.

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[1] Mott, N. F., *Engineering*, **165**, 16 (1948). Bueche, A. M., and White, A. V., *J. App. Phys.*, **27**, 980 (1956).

[2] Kies, J. A., Sullivan, A. M., and Irwin, G. R., *J. App. Phys.*, **21**, 716 (1950).

[3] Wolock, I., Conference on Fracture, Swampscott, Mass. (1959).

[4] Benbow, J. J., and Roesler, F. C., *Proc. Phys. Soc.*, **70**, 201 (1957). Berry, J. P. (unpublished observations).

[5] Bueche, A. M., and Berry, J. P., Conference on Fracture, Swampscott, Mass. (1959).

Flow of Electrolyte within a Propagating Stress-Corrosion Crack

THE observed corrosion potential at the mouth of a propagating crack in stressed Cu_3Au immersed in $FeCl_3$ is as much as 0·1 V. negative relative to that at uncracked parts of the surface[1]. This suggests that the advancing crack edge is dissolving anodically at an even lower potential. Potential measurements on stress-corroding wires of 18–8 steel in hot magnesium chloride solution[2,3], where the average corrosion potential during cracking is only about 0·06 V. above the reversible potential, suggest further that the potential at the advancing edges must in fact be very close to the reversible value. This led Hoar and West[4] to study the electrochemistry of 18–8 surfaces made to represent, on a macroscopic scale, the yielding material at such edges. Their experiments showed that dissolution of yielding 18–8 proceeds at the reversible potential provided that concentration over-potential is eliminated by flowing electrolyte past the surface at a suitable rate. It may be inferred that the advancing edge of a propagating stress-corrosion crack dissolves continuously into substantially virgin electrolyte, in view of the concomitant yawning.

This hypothesis has been tested by using two rather simplified crack models, and it has been found that, provided that the sides of the crack are strongly wetted, electrolyte is readily transferred from the crack mouth to the propagating edge, although in the models used some mixing with the crack electrolyte took place.

Two pieces of 'Perspex' apparatus were used:

(*a*) *Apparatus A* : a curved surface is allowed to bend away from a plane one, so that the 'crack' between them progressively yawns and is propagated at 0·5–10 cm./sec. This model most closely represents a true stress-corroding crack, although the 'profile ratio' (that is, crack-length divided by the amount of yawning at the mouth) is rather smaller (20 : 1) than that normally obtaining (nearer 100 : 1 for 18–8). The crack can be propagated a total distance of 5 cm., at which point it encounters a probe Ag,AgCl electrode. Provision is made for injecting chloride ions or a suitable dye into the electrolyte just in front of the crack mouth.

(*b*) *Apparatus B* : two straight surfaces are slid over one another at a rate of 0·2–5 cm./sec. The angle of contact, which determines the profile ratio, may be adjusted to give ratios between 15 : 1 and ∞ : 1 and the probe electrode is situated always at the advancing crack edge. Again, there is provision for injecting a 'marker' solution just in front of the crack mouth. The sliding action is thought to militate against indrawing of fresh electrolyte instantaneously to the crack edge, in view of the stirring produced, so that any positive results are probably conservative.

While neither apparatus gave rise to any sudden discontinuity in the potential of the probe electrode when saturated potassium chloride flowed past the mouth of a crack being propagated in tap water as electrolyte, measurable effects occurred when 'Teepol' was present. If we convert the observed potential-changes into changes in chloride ion concentration, apparatus *A* exhibited local concentration increases at the advancing edge of between 1·1 and 1·9 times. For apparatus *B*, where the average crack length was 4–5 cm. and the propagation-rate about 2 cm./sec. (that is, some 10^4 times the propagation-rate in true stress-corrosion cracks in 18–8), the local concentration change was 1·7–4·5 times. Oddly, the concentration rise at the crack edge in apparatus *B* did not seem unduly dependent upon profile ratios up to 100 : 1, although as expected a ratio of ∞ : 1 (that is, a closed crack) led to no rise. The addition of saturated potassium chloride directly to the vicinity of a similar electrode gave a local concentration rise of about 20 times.

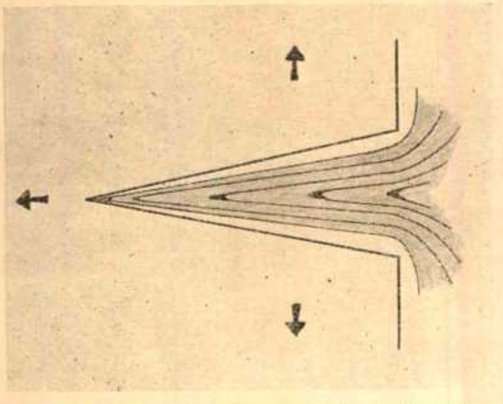

Fig. 1. Flow of electrolyte into propagating crack (with scale greatly exaggerated in the vertical direction)

With alcohol as electrolyte and deep red dye as 'marker', solution flow into a propagating crack was observed as in Fig. 1. So far as it was possible to tell, dye was conveyed instantaneously to the crack edge in apparatus *A* but results were less clear for apparatus *B*.

It may reasonably be concluded that, for long thin propagating cracks, anodic dissolution occurs into electrolyte contaminated no more than five times (the 'best' value from apparatus *B* was 20/4·5 = 4·4 times) by the previous crack contents, so long as the electrolyte thoroughly wets the sides of the crack. Owing to the inferred electrochemical nature of crack propagation in 18–8 steels[2,4], where capillarity must be such as to cause hot chloride solution to penetrate each crack right to the advancing edge, it is probable that wetting in this instance is similar to that which obtains for alcohol and aqueous solutions of 'Teepol' on surfaces of 'Perspex'.

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[1] Gerischer, H., and Rickert, H., *Z. Elektrochem.*, **46**, 681 (1955).

[2] Hoar, T. P., and Hines, J. G., *J. Iron and Steel Inst.*, **182**, 124 (1956).

[3] Hoar, T. P., and Hines, J. G., Int. Comm. for Electrochem., Thermodynamics and Kinetics (CITCE) VIII, 1956, 273 (Butterworths, London, 1958).

[4] Hoar, T. P., and West, J. M., *Nature*, **181**, 835 (1958).

GEOLOGY

Radiocarbon Measurements and Annual Rings in Cave Formations

The radiocarbon method has been used to test the hypothesis that the prominent rings found in certain calcite deposits of Moaning Cave in California represent annual growth variations. Carbon-14 analyses were made on two samples taken from a presumably active section of travertine covering a human femur (Fig. 1). When discovered in 1951, the specimen had 1,206 identifiable rings with an estimated 200 additional rings obscured[1]. Thus, if these rings formed annually and without interruption the bone at the centre became the nucleus of the cave formation about 1,400 years ago.

The locations of the two samples (*L*-530*A* and *L*-530*C*) taken for radiocarbon analysis are shown in Fig. 1. The two dotted lines overlie rings which on a geometrical basis have the mean age of the analysed material. In determining the position of these dotted lines a linear rate of growth is assumed. The radiocarbon concentration of the ring corresponding to the dotted line should thus be equal to the average radiocarbon concentration of the entire sample.

In Table 1 are the results of the radiocarbon analyses, expressed as fractions of the carbon-14/carbon-12 ratio in age-corrected pre-1900 woods (tree-ring samples corrected for decay of carbon-14 between the growth date and the present). The results in column 4 are uncorrected for isotope fractionation effects, while those in column 6 are normalized to a common carbon-13 concentration based on the estimated carbon-13 values given in column 5. The results in column 6 may be thought of as those which would have been obtained if isotope fractionation did not occur in Nature.

Table 1

Sample	Location in specimen (cm.)	Depth to mean age	Fraction pre-1900 wood $^{14}C/^{12}C$	δ ^{13}C* per mil	Normalized fraction pre-1900 wood $^{14}C/^{12}C$
L-530*A*	6·1–8·8	7·5	0·931 ± 0·017	(−5)	0·890 ± 0·017
L-530*C*	0·0–2·6	1·3	0·826 ± 0·010	(−5)	0·784 ± 0·010

* Relative to the Belemnite standard.

In computing ages from these results the main problem is that the initial carbon-14/carbon-12 ratio in the samples cannot be independently estimated. As shown by Deevey *et al.*[2], freshwater carbonates can have abnormally low carbon-14/carbon-12 ratios because of the incorporation of carbon derived from old limestones. In no case, however, could the samples, when deposited, have had carbon-14/carbon-12 ratios exceeding that in age-corrected pre-1900 wood. This means that upper limits can be set on the ages of the two samples by directly inserting into the carbon-14 age equation the ratios in column 6. The results are 950 years for sample *L*-530*A* and 1,950 years for sample *L*-530*C*.

Without even knowing the original carbon-14/carbon-12 ratio of the deposited carbonate, a more meaningful estimate can be made in the following way. If in the section considered the carbon-14/carbon-12 ratio in newly deposited calcite is assumed to remain constant with time, the difference between the radiocarbon concentration in the two samples is a direct indication of the average time intervening between their deposition. The calculations indicate that the 1·3-cm. layer was deposited 1,000 ± 200 years earlier than the 7·5-cm. layer. If the rate of accumulation between the 0·0- and the 1·3-cm. levels is assumed to be the same as the average rate over the 1·3–7·5-cm. span, the time required to deposit the first 1·3 cm. of calcite was 200 years. Similar reasoning yields 200 years for the time interval between the 7·5-cm. level and the surface of the specimen (8·8 cm.). Hence, based on the assumptions of constant initial carbon-14/carbon-12 ratio and of a linear growth-rate, the accumulation period for the entire 8·8 cm. of travertine is estimated as 1,400 ± 250 years. It should be noted that this conclusion is independent of the time at which deposition ceased.

The further assumption that the formation was actively growing when the specimen was collected makes possible an estimate of the initial carbon-14/carbon-12 ratio. In this event the 7·5-cm. level would have an age of 200 years and the 1·3-cm. level an age of 1,200 years. This assumption fixes the initial carbon-14/carbon-12 ratio of newly deposited carbonate at 0·915 that in age-corrected pre-1900 wood.

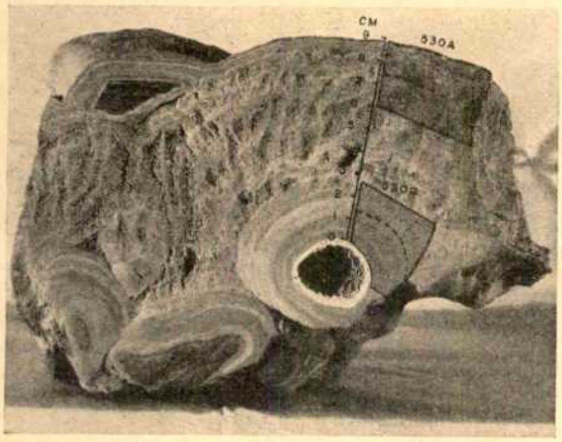

Fig. 1. Photograph of section cut through travertine deposit. The location of the samples analysed for carbon-14 are outlined. The age computed for the dotted mid-line for sample *L*-530*A* is 200 ± 100 and for the dotted mid-line of *L*-530*C* is 1,200 ± 200

It is of interest to compare this result with that obtained on a sample of recently formed calcite from Crystal Palace Cave located in Tuolume County, California. Supplied by R. de Saussure, this sample consisted of calcite covering and replacing a piece of wood found in the cave. A radiocarbon analysis of the wood yielded an age of less than 150 years and confirmed de Saussure's contention that it was a recent addition to the cave. The carbon-14/carbon-12 ratio for the calcite was 0·937 (± 0·008) that in age-corrected pre-1900 wood. When normalized for isotope fractionation, the result becomes 0·901 ± 0·008 (based on δ-carbon-13 = − 7·2‰)—in good agreement with the value of 0·915 required for the Moaning Cave samples.

In the literature, most of the published results for terrestrial waters are less than 0·90. For example, Deevey *et al.*[2] give values around 0·80 for alkaline lakes, while Brinkman, Munnich and Vogel[3] cite values of 0·66–0·84 for alkaline ground water. In addition, Broecker and Walton[4] give 0·67 as the average for the dissolved carbonate in rivers draining sedimentary areas. However, Broecker and Walton[4] find values of 0·90–0·98 for streams draining igneous areas devoid of carbonate rocks. This difference between 'igneous' and 'sedimentary' rivers arises from the fact that the bicarbonate in rivers draining only silicate rocks is almost entirely from soil carbon dioxide which has reacted with silicate minerals, whereas in carbonate areas the bicarbonate of the carbon is derived partly from soil carbon dioxide and partly from limestone free of carbon-14. (Soil carbon dioxide has a carbon-14/carbon-12 ratio approximately equal to that in contemporary wood.) Although the samples reported here are from limestone caves, the waters entering these caves apparently have derived nearly all their bicarbonate from soil carbon dioxide.

In conclusion, then, the radiocarbon results suggest that the rings observed in the travertine are annual, probably reflecting seasonal changes in the mode of deposition. This is consistent with Orr's[1] study of calcite coatings on nails which had remained in Moaning Cave for 29 years (1922–51). Using high magnification he found up to 29 rings. Whether this applies to cave formations in general awaits similar studies on other deposits. The present study merely demonstrates how the radiocarbon method can be applied to such problems.

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[1] Orr, Phil C., "Excavations in Moaning Cave, Santa Barbara", Museum of Natural History, Dept. Anthropol. Bull. No. 1 (1952).

[2] Deevey, jun., E. S., Gross, M. S., Hutchinson, G. E., and Kraybill, H. L., *Proc. U.S. Nat. Acad. Sci.*, **40**, 285 (1954).

[3] Brinkman, R., Munnich, K. O., and Vogel, J. C., *Naturwiss.*, **1**, S., 10 (1959).

[4] Broecker, W. S., and Walton, A., *Geochim. et Cosmochim. Acta*, **16**, 15 (1959).

CHEMISTRY

Substitution Reactions of Cobaltic Complexes catalysed by Adsorption

THE rate of the reduction of the complexes $[Co(NH_3)_5X]$ at the dropping mercury electrode depends on the nature of the supporting electrolyte and increases in the order: $SO_4^{=} \ll ClO_4^- < NO_3^- < Cl^- < Br^-$ as is manifested by the shift of the half-wave potential of the reduction wave with the change of the supporting electrolyte. The halides Cl^-, Br^- (and under suitable conditions also I^-) cause a further, quite different effect: at a certain concentration of the halide ion a new reduction wave appears which starts from the potential of the dissolution of mercury. The relative height of this wave (that is, the ratio of the positive wave to the total diffusion current) increases non-linearly with increasing concentration of the anion until all the complex is reduced in this positive wave (Fig. 1).

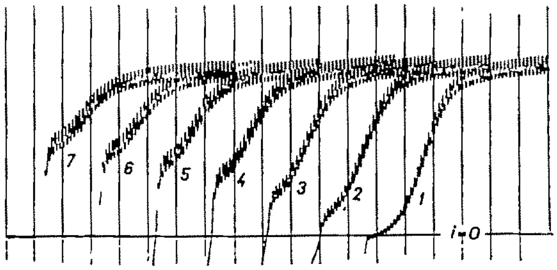

Fig 1. The influence of Br^- ions on the reduction wave of $[Co(NH_3)_5.H_2O]^{3+}$.
Solution. 0 1 *M* $HClO_4$, 0·9 *M* $NaClO_4$, 2×10^{-3} *M* $[Co(NH_3)_5H_2O].(ClO_4)_3$.
Curve 1, 0, 2, 2×10^{-4}; 3, 4×10^{-4}; 4, 6×10^{-4}; 5, 8×10^{-4}; 6, 10×10^{-4}; 7, 12×10^{-4} *M* NaBr.
100 mV./Absc, from −0·2 V. against mercurous sulphate electrode; *S* = 1 40

The concentration of the anion necessary for the development of the positive wave of given relative height depends on the nature of the anion: Cl^- ions have a much smaller effect than Br^- ions. Also the nature and concentration of other anions present in the solution influence the relative height of the positive wave, the latter being highest in solutions of pure halides and decreasing by addition of ClO_4^-, NO_3^- or $SO_4^{=}$ ions.

This behaviour suggests that the addition of halides changes the mechanism of the reduction of the cobaltic complex. This is further supported by the fact that the variation of the positive wave with the height of the mercury reservoir shows that, if the relative height of the first wave is small, the transport of the depolarizer to the electrode surface is governed not by diffusion but rather resembles that governed by a reaction preceding the electrode reaction proper. Furthermore, the properties of the positive wave are almost identical with those of the wave of $[Co(NH_3)_5Cl]^{2+}$ or $[Co(NH_3)_5Br]^{2+}$ (Vlček, A. A., unpublished results), so that these ions are probably the actual depolarizing particles. From this it could be concluded that in presence of halides the reaction:

$$[Co(NH_3)_5X]^{2+} + Cl^- \underset{k}{\rightarrow} [Co(NH_3)_5Cl]^{2+} + X^- \qquad (1)$$

(or similarly with Br^- or I^-) occurs in the solution surrounding the electrode and furnishes the actual depolarizer. Then the height of the first wave would be given by the rate of the homogeneous reaction (1). The value of the rate constant k measured by other methods[1] for the reaction of the $[Co(NH_3)_5H_2O]^{3+}$ with Cl^- ions is of the order of 10^{-5}(mol./l.)$^{-1}s^{-1}$, and it is thus impossible that the reaction in the solution near the electrode would contribute to the positive wave: the rate constant necessary for the observed currents would have to be at least 10^7 times higher than that measured in the solution.

To explain all the observed phenomena it has to be assumed that the particle actually exchanging the electrons with the electrode at the potential of the first wave is supplied by a chemical reaction proceeding directly at the electrode surface, the reactants being in the adsorbed state. This assumption that the reaction proceeds as a heterogeneous one is supported by the study of the current-time curves at constant potential. For small relative heights these curves obey the equation $i = \text{const.} \times t^{\alpha}$, where $\alpha > 2/3$, which is a property characteristic of processes in which absorption plays a decisive part[2]. The influence of surface-active substances, in the presence of which the relative height of the first wave is diminished, points to the same conclusion.

When a reaction proceeds directly at the electrode surface, all the particles formed react immediately with the electrode. On the other hand, the amount of particles formed per unit time in the adsorbed monomolecular layer is much smaller than that in the volume of the solution so that the assumption that the reaction proceeds in the adsorbed state would not explain the observed phenomena. Hence a further assumption has to be made, namely, that the reaction proceeds by a quite different rate, being catalysed by the adsorption at the surface of the electrode. Such catalysis of reactions of complexes has been observed in many cases (for review see ref. 3), the catalyst being charcoal, silica gel, mercury, Raney nickel, etc. The reaction which leads to the appearance of the positive wave is of a similar nature. It is a substitution reaction catalysed by the adsorption at the surface of the electrode.

The polarographic method can thus be used for the study of the ability of various complexes to undergo these heterogeneous substitutions, the relative height of the first wave being a measure of the rate constant of the reaction. For a series of complexes $[Co(NH_3)_5X]$ the following order of the reaction rates with Br^- ions in 0·1 *M* $HClO_4$, 0·9 *M* $NaClO_4$, 5×10^{-3} *M* NaBr solution has been deduced, X being:

$$NO_2^- < NH_3 \approx F^- < Ox^{=} \approx (ONO)^- < CH_3COO^- < H_2O < NO_3^- \qquad (2)$$

The adsorption catalysis is probably due to the strong polarization of the complex owing to the formation of a bond with the catalyst and to the influence of the electric field in the interface between the catalyst and the solution. The activation energy of a process is much decreased by such a polarization, the complex being much more reactive in the adsorbed state. This mechanism is identical with that which is supposed to operate in the electrode reactions[4], as can be seen also by the close parallelism between the rate of the catalysed substitution reactions and the rate of the electrode reactions; the order of the latter being in 1 *M* $NaHSO_4$ (ref. 4 and Vlček, A. A., unpublished results):

$$NH_3 < F^- < Ox^{=} < (ONO)^-, NO_2^- < CH_3COOH < H_2O < NO_3^- \qquad (3)$$

With the exception of the complex with nitrite, the two series are almost identical.

This close parallelism suggests that the structural changes of the reacting complexes, that is, the configuration of the activated complex, are very similar in both cases. For the electrode reaction it has been found[4] that, with the exception of the complex $[Co(NH_3)_5NO_2]^{2+}$ (Vlček, A. A., unpublished results), the complexes are orientated in such a way that the atomic group X is as distant as possible from the electrode surface. This orientation is also very convenient for the substitution reaction, the heterogroup being accessible for the exchange with other reactants without any bond breaking and rearrangement of the complex.

The proof of substitution reactions catalysed by adsorption at the electrode surface is very important for the interpretation of many observations concerning the influence of anions on the electrochemical behaviour of complexes. The presence of many anions causes a great shift of the reduction wave of some complexes towards positive potentials. This shift may be due to a substitution reaction of the type here described, which, when rapid, causes the reduction of the total amount of the depolarizer to take place at positive potentials.

The experimental details and further discussion will be published elsewhere.

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[1] Adell, B., *Z. anorg. Chem.*, 246, 303 (1941).

[2] Weber, J., Koutecký, J., and Koryta, J., *Z. Elektrochem.*, 63, 583 (1959).

[3] Basolo, F., and Pearson, R., "Mechanisms of Inorganic Reactions" (John Wiley and Sons, Inc., 1958)

[4] Vlček, A. A., *Discuss. Farad. Soc*, 26, 164 (1958).

Microwave Spectrum of Fluorine Cyanide

A fraction of the product of fluorination of cyanogen, in which fluorine cyanide was detected by infra-red spectroscopy[1], has now been shown to absorb in the microwave regions of 21,000, 42,000 and 63,000 Mc./s. The absorptions occur in the frequency-ranges expected from the resolved rotational structure of the infra-red band, ν_3, centred on 1,077 $cm.^{-1}$. Their Stark effects and nuclear quadrupole splittings are those expected for $J = 0 \rightarrow 1$, $1 \rightarrow 2$ and $2 \rightarrow 3$ transitions in the linear molecule, $FC^{14}N$. Spectra due to molecules with one quantum of vibrational energy in the degenerate bending mode (ν_2) have also been measured.

Values so far obtained are the rotational constant, $B_0 = 10{,}554{\cdot}20 \pm 0{\cdot}01$ Mc./s.; the coupling constant for $^{14}N = -2{\cdot}67 \pm 0{\cdot}05$ Mc./s.; the dipole moment, $\mu = 1{\cdot}68 \pm 0{\cdot}05\ D$; and the centrifugal stretching constant, $D_J = 0{\cdot}0053 \pm 0\ 0003$ Mc /s. The quadrupole coupling suggests a C—N distance slightly more than 1·163 A. found[2] in ClCN: if 1·165 A. is assumed, the C—F distance is obtained as 1·260 A. This is the shortest C—F distance known, and implies, in agreement with the low value of the coupling constant, an important contribution of $^{+}F{=}C{=}N^{-}$ to the structure.

We are at present attempting to improve the yield and the purity of the sample. It should then be possible to detect other isotopic species in natural abundance (particularly $FC^{15}N$). We hope to be able to detect and resolve other infra-red bands to see if the small discrepancy between the microwave and the infra-red (parallel mode) B-values extends to infra-red values obtained from perpendicular modes.

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[1] Aynsley, E. E, Dodd, R. E., and Little, R., *Proc. Chem. Soc.*, 265 (1959)

[2] Gordy, W., Smith, W. V., and Trambarulo, R F., "Microwave Spectroscopy", 371 (New York, 1953).

Thiol Exchange System with Thiolbenzoic Acid

Although many methods for the preparation of thiolacetic acid have been reported[1], all rely on the use of hydrogen sulphide, preferably under pressure, or phosphorous pentasulphide, which is not pleasant to handle.

It occurred to us that if an equilibrium existed between thiolbenzoic acid and thiolacetic acid:

$$C_6H_5COSH + CH_3COOH \rightleftharpoons C_6H_5COOH + CH_3COSH$$

the reaction might be persuaded to go towards completion by removal of the latter as the lowest-boiling component.

We have, in fact, obtained thiolacetic acid in yields of 95 per cent, based on the thiolbenzoic acid used, by this exchange method which we believe to be novel. Catalytic amounts of water appear to be essential; the most satisfactory method is to reflux glacial acetic acid (2 mol.) under a packed column fitted with reflux head, then gradually (8–12 hr.) add to the still, thiolbenzoic acid (1 mol.) previously saturated by shaking with water. The head temperature falls to 87–89° C. and the thiolacetic acid is withdrawn at such a rate that this temperature is maintained. The product needs drying for storage.

As thiolbenzoic acid is now commercially available the method should be attractive to those wishing to prepare conveniently relatively small quantities of thiolacetic acid.

Thiolpropionic acid was prepared by the same method, but attempts to convert butyl alcohol to butyl mercaptan were unsuccessful.

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[1] "Organic Syntheses", 31, 108 (1951).

RADIOCHEMISTRY

An Alternative Mechanism for Chemical Protection against Radiation Damage

Studies of the effects of chemicals in decreasing the radiation damage suffered by exposed organisms have revealed a number of compounds which are quite efficient in this respect[1-3]. While several mechanisms have been suggested for their mode of action[4], none has been universally accepted. It seems possible that the protective action may function through several alternative routes. It is the purpose of this communication to show another possible mode of action of these agents and some of the evidence which supports it. The mode of protection cited here need not be an exclusive one.

Almost all the best protective agents have structures characteristic of good chelating agents. The general structures of many of these agents can be written as $X—CH_2—CH_2—Y$ or $X—CH_2—CH_2—CH_2—Y$, where X and Y are groups capable of co-ordinating with one or more of the co-ordination sites of a metal ion. Thus these are compounds which can readily form five- or six-membered rings upon chelation. Typical examples of such compounds are β-mercaptoethylamine, glutathione, 2,2′-dithioethanol, 3-mercaptopropylamine and S,2-aminoethyl*iso*thiourea. It is suggested that their effectiveness has the following basis. In the course of radiation some of the transition metal constituents of the organism are ejected from their normal sites by the radiation. These are then more or less free to enter the circulation and do so in an amount sufficiently large to poison many of the metal ion-inhibited enzymes such as adenosine triphosphatase, carbonic anhydrase, arginase, etc.[5]. In the presence of the protective agent the 'outlaw' metal ions are tied up in the form of chelates which are more stable than the complexes formed with the enzyme. Thus no inhibition of such enzymatic processes can occur. Since many such inhibition processes are irreversible, the ineffectiveness of administering the protective agent after irradiation is explicable.

If such is the case, a correlation between the stability of the resulting chelate and the effectiveness of the protection furnished would be expected. Unfortunately, quantitative studies of both stability constants and survival rates are available on only a few compounds. For the compounds β-mercaptoethylamine, cystein, histamine, ethylenediamine tetraacetic acid, ethylenediamine, salicylic acid, and glycine such data are available. The correlation coefficient between the percentage survival[1-3] and the $\log_{10}$ of the stability constants of the copper chelates[6] is 0·57. For a perfect correlation this would be 1·0 and for no correlation it would be 0.

If this mechanism is actually responsible for a considerable part of the protective action, it would suggest that the best protection would be afforded by compounds capable of tying up several of the co-ordination positions of a metal ion and thus use the chelate effect to enhance the protective action. The same substances which provide chemical protection should also provide protection against heavy-metal poisoning when injected before the heavy-metal. Furthermore, a wider class of materials should be capable of protective action than would be estimated on the basis of other theories of this action, such as that of mixed disulphide formation[7]. The requirements of a good protective agent appear to be very similar to those of a good chelating agent with some specificity for transition metals, especially copper. If this chelating action is important, the protective action found for ethylenediamine tetraacetic acid[3,8] should be found in enhanced measure with compounds such as 1,2-diamino*cyclo*hexane N,N,N′,N′-tetraacetic acid and diethylenetriaminepentaacetic acid.

An alternative explanation with somewhat similar consequences is one in which the catalytic effectiveness of the resulting chelate complexes in decomposing hydroxylonium or hydrogen peroxide is the basis of their action. This explanation could perhaps be reconciled with the theory of competition for hydroxylonium radicals of Bacq *et al.*[1,3].

Bacq and his co-workers briefly considered the possible importance of chelation but then dismissed it. Their chief evidence against it was obtained not on organisms but on polymethacrylic acid solutions, where the action of radiation is presumably analogous to that found with organisms. It seems very likely that the behaviour of organisms is considerably more complex. It is highly possible that there are two or more major types of radiation damage the cumulative effect of which is lethal. The control or elimination of one of these types may then reduce the cumulative damage to a level which is not lethal.

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[1] Bacq, Z. M., and Alexander, P., "Fundamentals of Radiobiology", Chap. 14 (Academic Press, Inc., New York, 1955).

[2] Doherty, D. G., Burnett, jun., W. T., and Shapira, R., *Radiation Res.*, 7, 13, 22 (1957).

[3] Alexander, P., *et al.*, *Radiation Res.*, 2, 392 (1955).

[4] Pihl, A., and Eldjarn, L., *Pharmacol. Rev.*, 10, 437 (1958).

[5] Laidler, K. J., "Introduction to the Chemistry of the Enzymes" (McGraw-Hill Book Co., Inc., New York, 1954).

[6] Bjerrum, J., Schwarzenbach, G., and Sillen, L. G., "Stability Constants", Pt. 1 (Chemical Society, London, 1957).

[7] Eldjarn, L., and Pihl, A., *Radiation Res.*, 9, 110 (1958).

[8] Bacq, Z. M., Herve, A., and Fischer, P , *Bull. Acad. Med. Belg.*, 6th series, 18, 226 (1953).

BIOCHEMISTRY

Incorporation of α,ω-Dicarboxylic Acids as Side-chains into the Penicillin Molecule

WHILE a considerable number of biosynthetic penicillins have been obtained by adding different side-chain precursors to the culture medium, the incorporation of aliphatic dicarboxylic acids into the penicillin molecule does not as yet seem to have been reported.

When a high-yielding *Penicillium chrysogenum* strain of the Wisconsin family (*Wis* 51.20 [1], subculture *c* isolated in this laboratory) was grown in shake flasks on a corn-steep lactose medium in the absence of a precursor acid (for conditions of growth see footnote of Table 1), the culture medium showed a slight activity against *Bacillus subtilis* (40–150 benzylpenicillin units per ml.) and a slightly higher activity (about 300 units per ml., benzylpenicillin as standard) against *Klebsiella pneumoniae*. Addition of the lower homologues of α,ω-dicarboxylic acids gave essentially similar results ; but on addition of adipic acid or the higher homologues the antibacterial activity against *K. pneumoniae* considerably increased while that against *B. subtilis* was slightly decreased (Table 1).

The highest increases of anti-*Klebsiella* titres (up to ten-fold) were obtained on addition of adipic, pimelic and azelaic acid ; the higher homologues gave somewhat lower titres. Similar results were obtained on a synthetic medium of the type developed by Jarvis and Johnson[2].

The penicillin formed after addition of adipic acid as side-chain precursor was isolated from larger quantities of culture media in purified form and

Table 1. ANTIBACTERIAL ACTIVITY AGAINST *B. subtilis* AND *K. pneumoniae* OF CULTURE FILTRATES OF *P. chrysogenum* STRAIN *Wis* 51.20 c AFTER 96 HR.

Precursor acid added		Final concentration in percentage of free acid‡	Activity against* (benzylpenicillin units/ml.) *B. subtilis*	*K. pneumoniae*†
None			185	280
Phenylacetic	(sodium salt)	0·2	1,700	1,600
Oxalic	(potassium salt)	0·7	200	330
Malonic	(sodium salt)	0·6	175	340
Succinic	,,	0·65	170	290
Glutaric	,,	0·7	170	300
Adipic	,,	0·8	100	1,000
Pimelic	,,	0·9	130	1,260
Suberic	(potassium salt)	0 95	150	1,040
Azelaic	,,	1·0	110	1,030
Sebacic	,,	1·1	100	900
1,10-decanedicarboxylic (C_{12})	,,	1·25	100	650
1,12-dodecanedicarboxylic (C_{14})	,,	1·4	100	800

Conditions of growth: Culture medium—lactose 3 per cent, glucose 0·5 per cent, corn-steep liquor 4 per cent, calcium carbonate 1 per cent, seeded with 10 per cent of a 48-hr.-old mycelial suspension grown on a corn-steep liquor (8 per cent) – dextrin (6 per cent) medium, *p*H adjusted to 5 4 with sodium hydroxide, from a suspension of conidia developed on barley moistened with the sporulating solution of Whiffen and Savage (ref. 4). 500-ml. Erlenmeyer shake flasks with 60 ml. of culture media were agitated at 25° C. on a rotary shaker (ref. 9), sulphite aeration rate 35 ml. oxygen per 100 ml. fluid per hr.

* Averages of two shake flasks which gave results in very good agreement with each other.

† The figures are benzylpenicillin units giving inhibition zones equal to those given by the undiluted broth.

‡ Added in two lots of 1–4 ml. each (according to the solubility), at 24 hr. and 72 hr., of an aqueous solution of the sodium or potassium salts in concentrations to give the above final concentration values

shown to be (4-carboxy-*n*-butyl)-penicillin, that is, to contain the adipic acid side-chain.

The mycelium was grown at 28° C. in fermentors of 90 l. capacity[3] containing 50 l. of a culture medium consisting of 4 per cent corn-steep liquor, 3 per cent lactose, 0·5 per cent glucose and 1 per cent calcium carbonate. (Conditions of aeration and agitation: sparger aeration in fully baffled system, propeller blade 130 mm. diameter rotating at 420 r.p.m., air flow 50 l. per min. during first 24 hr., then increased to 80 l., overpressure 1 atm. One portion of 50 ml. of vegetable (mainly soy bean) antifoam oil containing 4 per cent 'Alkaterge', added at the beginning, and further portions of 50 ml. during the fermentation if and when required.) The culture medium was seeded with 10 per cent of a 48-hr.-old mycelial suspension grown in another 90-l. fermentor under the same conditions of aeration in a medium containing 6 per cent dextrin, and 8 per cent corn-steep liquor, *p*H adjusted to 5·4 with sodium hydroxide. The seed medium was inoculated with a suspension of conidia, developed on barley moistened with the sporulating solution of Whiffen and Savage[4].

Adipic acid in the form of its sodium salt was added in a 25 per cent aqueous solution in two lots of 800 ml., one at 24 hr. and the second at 72 hr., to give a final concentration of 0·4 per cent after each addition, that is, a total of 0·8 per cent. The fermentation was allowed to proceed for 96–120 hr., after which time the titre against *K. pneumoniae* had reached about 1,500 benzylpenicillin units/ml., while the titre against *B. subtilis* was about 150 units/ml. The filtered culture medium was percolated through a column of granular charcoal (20–60 mesh, *BO* grade, Farnell Carbons, Ltd., London) prepared as described by Abraham, Newton and Hale[5]. 1 vol. of charcoal was used for 2 vol. of medium. After washing with 1/10 vol. of water and then 1 vol. of 20 per cent acetone, the active substance was eluted with 2 vol. of 70 per cent acetone. The eluate containing about 70 per cent of the active substance with a specific activity of about 80 hydroxylamine units/mgm. using the method of Ford[6] for the determination of penicillin (the value of the pure compound is 1,500 units/mgm.) was concentrated about 20 times *in vacuo* in a climbing film still[7] at a temperature not above 40° C., and then lyophilized. There was a loss of activity of about 20 per cent in this process. The dry material from the lyophilization was extracted 3 times with 4 ml. of methanol for each gram of dry substance. 2 vol. of acetone were added to the methanolic solution and the mixture was centrifuged to eliminate precipitated material of low activity. The supernatant was concentrated to a small volume and 10 vol. of acetone were added to this. The supernatant contained a mixture of penicillins (about 40 per cent of the total activity) with a ratio of activity against *K. pneumoniae*/activity against *B. subtilis*, similar to that of benzylpenicillin, while the precipitate contained the remainder of the active substance with a much higher activity ratio and a specific activity of 300–350 hydroxylamine units per mgm. The precipitate dissolved in water was passed through a column of 'Dowex' 1 × 8, 50–100 mesh, in the chloride form. 1 ml. of resin was used for about 10,000 hydroxylamine units of active substance. The activity was eluted with 5 per cent sodium chloride at *p*H 7. The active fractions were lyophilized, and the dry product was extracted with three portions of 100 ml. of dry ethanol. The addition of 4 vol. of acetone to the ethanolic solution precipitated a product which was dried with acetone and ether.

The yield from the ion-exchange chromatography and the successive operations was about 50–60 per cent and the final product was 70–80 per cent pure. The active product was rapidly destroyed by penicillinase. It could not be extracted by any organic solvents at an acid *p*H. In paper chromatography on Whatman No. 1 at 5° C., in *n*-butanol/ethanol/water, 40 : 10 : 50 v/v/v, it remained at the origin; in 70 per cent aqueous *n*-propanol it had an $R_F = 0{\cdot}35$ while benzylpenicillin had an R_F of 0·75. On paper electrophoresis (0·1 *M* phosphate buffer, *p*H 6·8, applied potential 25 V./cm.) it had a mobility about twice (20 cm. per hr.) that of benzylpenicillin (10·5 cm. per hr.). After hydrolysis for 2 hr. at 100° C. in 2·5 *N* hydrochloric acid, subsequent ether extraction and evaporation of the ether extracts, crystals appeared in a yield of 60 per cent of the theoretical

Table 2. COMPARISON OF ANTIBACTERIAL PROPERTIES OF BENZYLPENICILLIN AND (4-CARBOXY-*n*-BUTYL)-PENICILLIN*

	Benzylpenicillin (μgm./ml.)	4-Carboxy-*n*-butyl penicillin (μgm /ml.)†
Bacillus subtilis	0 027	0 52
Micrococcus flavus	0 30	1·57
Sarcina lutea	0·01	0 65
Staphylococcus aureus (Oxford)	0 06	1·57
Streptococcus pyogenes	<0·4	>32
Klebsiella pneumoniae	18	18
Salmonella typhimurium	10 84	4
Salmonella typhi	2 4	5
Escherichia coli	36·14	>67

* Serial broth dilution technique: 0·1 ml. of a 1 : 80 dilution of a 24-hr. broth culture of the respective bacteria was inoculated into 9 9 ml. broth of the following composition—meat extract 1 0 per cent, sodium chloride 0 5 per cent, peptone 1·0 per cent, glucose 0·05 per cent.

† Corrected for impurities.

amount expected, which were identified as adipic acid by melting and mixed melting point as well as by their paper chromatographic behaviour in two solvents (ethanol/conc. ammonia/water, 80 : 4 : 16, v/v/v, and *n*-butanol/acetic acid/water, 40 : 10 : 50, v/v/v).

A comparison of the minimal growth-inhibiting concentrations of (4-carboxy-*n*-butyl)-penicillin and benzylpenicillin against a range of different Gram-positive and Gram-negative bacteria is given in Table 2. From the results it is clear that (4-carboxy-*n*-butyl)-penicillin is very similar in its activity to cephalosporin *N* as reported by Heatley and Florey[8], that is, it has a much lower activity than benzylpenicillin against the Gram-positive and about the same order of activity against the Gram-negative bacteria. The lowering of the activity against Gram-positive bacteria is therefore not a specific effect of the α-amino group in the α-aminoadipic side-chain, present in cephalosporin *N*, but can be obtained by the introduction of the simple adipic acid side-chain or its higher homologues into the penicillin molecule, that is, by the presence of an extra free carboxylic group. A *K. pneumoniae* strain made resistant to benzylpenicillin by subculturing in the presence of subminimal inhibitory concentrations of benzylpenicillin (it grew eventually in 6,000 units per ml.) was also found to be resistant to (4-carboxy-*n*-butyl)-penicillin. Skin tests of (4-carboxy-*n*-butyl)-penicillin in seven highly benzylpenicillin-allergic subjects (carried out by Drs. R. Maffei and E. Errigo of the University of Rome) have shown that the compound does not elicit allergic reactions.

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[1] Backus, M. P., and Stauffer, J. F., *Mycologia*, 47, 429 (1955).
[2] Jarvis, F. G., and Johnson, M. J., *J. Amer. Chem. Soc.*, 69, 3010 (1947).
[3] Paladino, S., Ugolini, F., and Chain, E. B., *Rend. Ist. Sup. Sanità* (English edition), 17, 87 (1954).
[4] Whiffen, A. J., and Savage, G. M., *J. Bact.*, 53, 231 (1947).
[5] Abraham, E. P., Newton, G. G. F., and Hale, C. W., *Biochem. J.*, 58, 94 (1954).
[6] Ford, J. H., *Indust. Eng. Chem.*, Anal. Edit., 19, 1004 (1947).
[7] Van Heyningen, W. E., *Brit. J. Exp. Path.*, 30, 302 (1949).
[8] Heatley, N. G., and Florey, H. W., *Brit. J. Pharmacol.*, 8, 252 (1953).
[9] Paladino, S., *Rend. Ist. Sup. Sanità* (English edition), 17, 145 (1954).

Effect of Cardiac Glycosides and Inorganic Ions on Binding of Serotonin by Platelets

THE presence of high concentrations of serotonin (5-hydroxytryptamine) in blood platelets has stimulated many studies on the mechanism of the uptake of the amine by these cells[1-5]. This process can readily be studied *in vitro* employing platelets suspended in plasma or in saline solutions, and there is now considerable evidence that this process is one of active transport[6].

A previous study from this laboratory[7] employing platelets suspended in isotonic sodium chloride indicated that inorganic phosphate is required in order to obtain maximum binding of 5-hydroxytryptamine. This communication presents a more detailed study of the factors affecting the binding of 5-hydroxytryptamine by blood platelets in well-defined media. By controlling the *p*H, it appears that the active transport mechanism can be studied independently of other mechanisms that may be functioning. In such a system inorganic phosphate and potassium are required for maximal uptake. The cardiac glycosides are known to inhibit the active transport of cations by several tissues[8] as well as the uptake of iodide by isolated thyroid slices[9], and it seemed of interest to test their ability to inhibit the transport of other substances. Both the cardiac glycosides and reserpine, which has previously been shown to inhibit the uptake of 5-hydroxytryptamine by platelets[4], inhibited this system.

Human platelets were isolated according to the method of Dillard *et al.*[10], washed as described previously[7] and resuspended in saline. When platelets were incubated with 5-hydroxytryptamine the concentration of the latter in the platelets continued to increase for approximately 90 min. Incubations were carried out for 1 hr. at 37° C., during which time uptake was nearly linear. The platelets were re-isolated, washed and assayed for both protein[11] and 5-hydroxytryptamine[12]. Results are expressed as μgm. of 5-hydroxytryptamine per mgm. of platelet protein.

A marked effect of *p*H on the relationship between the uptake of 5-hydroxytryptamine and its concentration in the medium was observed. At *p*H 5·7 with potassium and phosphate present (Fig. 1) the system became saturated at low concentrations of amine (less than 5 μgm./ml.). At higher *p*H, however, another mechanism appeared, involving a more nearly linear dependence of 5-hydroxytryptamine accumula-

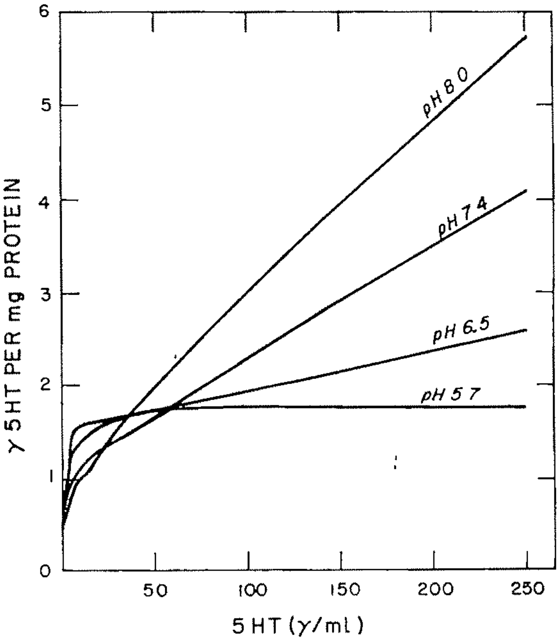

Fig. 1. The effect of *p*H and 5-hydroxytryptamine concentration on platelet uptake of the latter. The incubation mixture contained 1 ml. of platelet suspension (about 1 mgm. protein) in 0·9 per cent sodium chloride, 5-hydroxytryptamine (dissolved in 0·9 per cent sodium chloride and adjusted to *p*H 5·7), 50 μmoles phosphate buffered at the appropriate *p*H, and 0·9 per cent sodium chloride to make up to a final volume of 2 ml.

Table 1. EFFECT OF POTASSIUM AND PHOSPHATE ON UPTAKE OF 5-HYDROXYTRYPTAMINE AT pH 5 7
(μgm. of 5HT per mgm. of platelet protein over control*)

System	Experiment No. (μgm./mgm. protein) 1	2	3	4	Average percentage of maximal stimulation (γ)
A Platelet + 5HT + K^+ (45 μmoles) + PO_4^{3-} (50 μmoles)	0 62	0 84	1 17	0 62	100
B Platelet + 5HT + PO_4^{3-} (50 μmoles)	0 22	0 40	0·51	0·34	46
C Platelet + 5HT + K^+ (50 μmoles)	0·43	0 49	0 76	0 48	68

* Values are those over a control in which platelets were incubated only with 5HT (30 μgm) and 0 9 per cent sodium chloride to make up to a final volume of 2 ml

The incubations contained 1 ml. of platelet suspension in 0·9 per cent sodium chloride (about 1 mgm. protein), 30 μgm. 5HT (0·3 ml.) in 0 9 per cent sodium chloride and 0·9 per cent sodium chloride to make up to a final volume of 2 ml. 0 1 ml. of the following solutions was added System *A*, phosphate buffer pH 5·7 (0 5 *M* with respect to phosphate containing 450 μmoles/ml. of K^+ and 90 μmoles/ml. Na^+). System *B*, 0 5 *M* sodium dihydrogen phosphate (adjusted to pH 5·7). System *C*, 0 5 *M* potassium chloride.

5HT = 5-hydroxytryptamine.

tion on the amine concentration. This relationship between uptake and concentration suggests that a diffusion mechanism predominates at the higher pH value.

Platelets suspended in saline alone, at pH 5·7, were able to bind exogenous 5-hydroxytryptamine. However, the uptake was much greater when potassium and phosphate were added. As shown in Table 1 both phosphate and potassium were required for optimal uptake. Of the total attainable stimulation of 5-hydroxytryptamine uptake approximately 45 per cent could be produced by addition of phosphate alone and 70 per cent by potassium alone. Maximal effects were obtained with as little as 10 μmoles potassium and 1 μmole phosphate.

As shown in Table 2 both reserpine and oubain inhibited the uptake of 5-hydroxytryptamine in this system. However, the inhibition observed was only on the potassium and phosphate effects. The uptake of 5-hydroxytryptamine which occurred when the platelets were suspended in saline alone (usually a 2–3 fold increase over the control) was not affected by these drugs. The cardiac glycosides used routinely were oubain, digitoxin and strophanthin potassium, all of which had equal activity at 10^{-5} *M*. None of the glycosides inhibited at 10^{-7} *M*.

Table 2. EFFECT OF COMPOUNDS ON PLATELET UPTAKE OF 5-HYDROXY TRYPTAMINE AT pH 5 7
(Increase in μgm. of 5HT per mgm. of platelet protein over endogenous*

System	Experiment No. 1	2	3	4	5	6
A Platelet + 5HT	0 58	0·97	0 74	0 52	0·84	0 53
B Platelet + 5HT + K^+ + PO_4^{3-}	0·96	1·36	1·22	1·03	1·09	1·62
C Platelet + 5HT + K^+ + PO_4^{3-} + oubain	0·65	1 12	0·91	0·46	0 78	0 87
D Platelet + 5HT + K^+ + PO_4^{3-} + reserpine	0 53	1·08	—	0 52	0 70	0 60
E Platelet + 5HT + oubain	0·64	1·18	0 78	—	0·84	0 61
F Platelet + 5HT + reserpine	0 69	1 10	0·63	—	0·92	0 42

*The endogenous values in the experiments to be described in this report varied generally between 0 1–0·4 μgm. 5HT per mgm. of platelet protein.

Each incubation mixture contained 1 ml. of platelet suspension (about 1 mgm. of protein) and 30 μgm. of 5HT (0·3 ml). 50 μmoles of phosphate, 45 μmoles of K^+, oubain (10^{-4} *M* final concentration), and reserpine (10^{-5} *M* final concentration) were added where indicated. The final volume was made up to 2 ml. with 0 9 per cent sodium chloride.

5HT = 5-hydroxytryptamine.

Unlike the seroids and reserpine, 10^{-3} *M* fluoride inhibited almost completely the uptake of 5-hydroxytryptamine whether or not potassium and phosphate were present. Since the uptake of 5-hydroxytryptamine by the platelets proceeds under anaerobic conditions and is not inhibited by 2,4-dinitrophenol, it has been suggested that glycolysis supplies the energy required for this process. The inhibition observed with fluoride supports this postulate. The mechanism of inhibition by the glycosides remains unclear. However, in mammalian erythrocytes, which presumably derive energy for the transport of cations from glycolysis, the cardiac glycosides are also effective inhibitors of the transport of sodium and potassium[13] but do not affect the synthesis of adenosine triphosphate or the metabolism of the phosphorylated intermediates in glycolysis[14,15].

It is of interest that under more alkaline conditions (pH 8) where a diffusion type mechanism appears to predominate, reserpine, the cardiac glycosides and fluoride had much less effect on the accumulation of 5-hydroxytryptamine by platelets.

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[1] Humphrey, J. H., and Toh, C C., *J. Physiol*, 124, 300 (1954)
[2] Hardisty, R. M., and Stacey, R. S , *J. Physiol.*, 130, 711 (1955).
[3] Brodie, B. B., Tomich, E. G., Kuntzman, R., and Shore, P. A., *J. Pharmacol. Exp. Therap* , 119, 461 (1957).
[4] Hughes, F. B., Shore, P. A., and Brodie, B. B., *Experientia*, 14, 178 (1958).
[5] Born, G V. R., and Gillson, R E., *J. Physiol.*, 146, 472 (1959).
[6] Hughes, F. B., and Brodie, B B., *J. Pharmacol. Exp. Therap.* (in the press).
[7] Weissbach, H., Bogdanski, D. F., and Udenfriend, S., *Arch. Biochem. Biophys.*, 73, 492 (1958).
[8] Hajdu, S., and Leonard, E , *Pharmacol. Rev.*, 11, 173 (1959).
[9] Wolff, J., and Maurey, J. R , *Nature*, 182, 957 (1958).
[10] Dillard, G. H. L., Brecher, G , and Cronkite, E. P., *Proc. Soc. Exp. Biol. Med* , 78, 796 (1951)
[11] Sutherland, E. W., Cori, C. W , Haynes, R , and Olsen, N. S., *J. Biol. Chem.*, 180, 825 (1949).
[12] Weissbach, H . Waalkes, T. P., and Udenfriend, S., *J. Biol. Chem.*, 230, 865 (1958).
[13] Kahn, jun., J. B., and Acheson, G. H., *J. Pharmacol. Exp. Therap.*, 115, 305 (1955).
[14] Kunz, H., and Sulser, F., *Experientia*, 13, 365 (1957).
[15] Whittam, R., *J. Physiol.*, 140, 479 (1958).

Effect of Streptomycin on the Incorporation of Tyrosine labelled with Carbon-14 into Protein of *Mycobacterium* Cell Fractions *in vivo*

STREPTOMYCIN inhibits the incorporation of tyrosine labelled with carbon-14 into the protein of a cell-free system isolated from *Mycobacterium friburgensis*, but the degree of inhibition is less when intact cells are incubated in a culture medium[1].

We assume that streptomycin does not inhibit the incorporation into the protein of all cell fractions, and investigated, therefore, the rate of labelling of whole cell protein and various cell fractions on incubation of intact cells with labelled tyrosine.

The bacteria were washed and resuspended in a Sauton medium at a concentration of 200 mgm. wet

Table 1. EFFECT OF STREPTOMYCIN ON THE INCORPORATION OF LABELLED TYROSINE INTO PROTEIN OF *Mycobacterium friburgensis* CELL FRACTIONS *in vivo*

Experiment No.	Incubation time (min.)	c.p.m./mgm. protein							
		Whole cells		Fraction *A*		Fraction *B*		Fraction *C*	
		–	+ Streptomycin	–	+ Streptomycin	–	+ Streptomycin	–	+ Streptomycin
1	30	219	165	132	128	192	134	214	102
2	10	122	68	65	65	12	8	126	31
2	30	150	80	112	122	35	15	139	60
3	10	126	55	26	29	55	9	27	8

weight per ml. 5 ml. cell suspension were incubated for 30 min. with shaking at 37° C. in the presence of 5 μc./ml. labelled DL-tyrosine (1 μc. = 17 × 10^6 c.p.m.) with or without addition of 100 μgm./ml. streptomycin. The cells were washed twice with 20 vol. of ice-cold distilled water, and resuspended in 6 ml. buffer[2]. 1 ml. was removed to determine incorporation into total cell protein, to the remaining 5 ml. cell suspension 0·8 gm. alumina was added, and the cells were disintegrated by grinding in a chilled mortar for 20 min. The suspension was centrifuged at 1,200*g* for 10 min. (the sediment-fraction *A*). The supernatant was centrifuged in a Spinco preparative centrifuge at 105,000*g* for 60 min. to yield a particle fraction (*B*) and a clear supernatant (*C*).

The protein of the whole cells and of the fractions *A*, *B* and *C* was isolated and the radioactivity determined. The results of the experiments are shown (Table 1).

They show that the extent of inhibition differs markedly in the fractions, being greatest in the fraction *C* (which corresponds to our 'cell-free' system)[1], while no inhibition was found in fraction *A* (cell wall and membrane fragments).

These results suggest that streptomycin does not inhibit the protein synthesis of the cell wall.

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[1] Erdős, T., and Ullmann, A., *Nature*, **183**, 618 (1959).

[2] Keller, E. B., and Zamecnik, P. C., *J. Biol. Chem.*, **221**, 45 (1956).

Peptides obtained from Partial Hydrolysis of Decalcified Human Dentin Collagen

FROM *in vitro* studies of mineralizing connective tissues, it has been suggested that the arrangement of polarized protein macromolecules in the submicroscopic collagen fibrils may be an important factor in determining whether suitable 'nucleation centres' for hydroxyapatite crystals will be present[1]. The banded regions of natural and reconstituted collagens are thought to contain relatively high proportions of the bulky basic and acidic amino-acid residues, thus suggesting that the amino-acid residues in collagen are not randomly distributed. This view has already been verified for soft tissue procollagen by an analysis of peptides obtained by enzymatic hydrolysis[2]. Since soft and hard tissue collagens differ markedly from each other with respect to the availability of their ε-amino-groups to dinitrofluorobenzene[3], as well as in their solubility behaviour[1] and internal molecular structure[4], it is of interest to compare the composition, amino-acid sequence and structure of highly polar peptides obtained from both types of collagen.

Small pieces of dentin from the roots of freshly extracted adult human anterior teeth were demineralized by soaking in 1 *N* hydrochloric acid for 24 hr. at 4° C. The demineralized dentin collagen was washed with cold 0·16 *N* potassium chloride solution for 24 hr., rinsed with distilled water, and allowed to dry in air. Weighed samples (0·1 gm.) were heated in distilled water (20 ml.) in a sealed tube at 110° C. for 16 hr. The resulting hydrolysate was evaporated to small volume on a steam bath, and then separated into 4 ninhydrin-positive bands labelled *A*, *B*, *C* and *D*, by low-voltage electrophoresis for 2 hr. on Whatman No. 1 filter paper using 1·2 *N* acetic acid as solvent. Migration occurred towards the cathode. The positions of the amino-acids and peptide spots were visualized by spraying the dried chromatograms with 0·1 per cent ninhydrin in acetone, and heating at 105° C. for 2 min. Each band was eluted with 6 *N* hydrochloric acid (redistilled over stannic chloride), and hydrolysed for 16 hr. in a sealed tube at 105° C. Portions of the hydrolysate, as well as the mixture before acid hydrolysis, were examined by two systems of paper chromatography, employing butanol/acetic acid/water (4 : 1 : 5) and phenol/water (7 : 3) as solvents.

It was observed that the main products of the water hydrolysis of dentin collagen were peptides *A*, *B*, *C* and *D* together with minimal amounts of free amino-acids. The peptides *A*, *B*, *C* and *D*, labelled in order of increasing mobility towards the cathode, differed markedly from each other in their qualitative amino-acid composition. Band *B* was typical of the composition of the intact protein, all the amino-acids normally found in mammalian collagen being present. Band *A* resembled band *B*, except that hydroxyproline was absent. In band *C*, hydroxyproline, proline and phenylalanine were all absent. Band *D* contained 7 amino-acids—lysine, hydroxylysine, glycine, alanine, arginine, glutamic and aspartic acids. This highly polar band was further investigated and found to be homogeneous in the chromatographic systems used, and on electrophoresis at different *p*H values. By means of Sanger's dinitrofluorobenzene technique[5] it was shown that alanine was in the N-terminal position of the intact peptide *D*. Under the experimental conditions cited, the results were reproducible.

These preliminary results show that the distribution of amino-acids in dentin collagen is non-random and resembles to some extent that found in soft tissue procollagen[2]. The amino-acid sequence of the peptides, as well as their relative efficiency as 'seeding' agents for hydroxyapatite crystals, are currently being investigated, and the results will be reported at a later date.

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[1] Glimcher, M. J., *Rev. Mod. Phys.*, 31, 359 (1959).

[2] Solomons, C. C., and Irving, J. T., *Biochem. J.*, 68, 499 (1957).

[3] Grasmann, W., Hannig, K., Endres, H., and Riedel, A., *Hoppe-Seylers Z. physiol. Chem.*, 306, 123 (1956).

[4] Mechanic, G. L., and Levy, M., *J. Amer. Chem. Soc.*, 81, 1889 (1959).

[5] Sanger, F., *Biochem. J.*, 39, 507 (1945).

Inactivation of Erythropoietin by Neuraminidase and by Mild Substitution Reactions

ERYTHROPOIETIC activity in the plasma of rabbits made anæmic with phenylhydrazine is associated with mucoproteins[1–4]. It has been shown that the activity is destroyed by proteolytic enzymes[1,3,5]. Rambach *et al.*[6] have found as well as us that heating at 100°, *p*H 2, for 1 hr. destroys the activity, a treatment which, in the case of guinea pig α_1-acid glycoprotein[7], selectively removed sialic acid. In spite of the mildness of this treatment it is possible that other bonds (sugar, amino-acid) were broken. We have now been able to show that exposure to neuraminidase[8] completely inactivates the mucoprotein.

An acid glycoprotein (15·6 per cent sialic acid) has been regarded as erythropoietin by Rambach *et al.* We[2] have pointed out that it is probable that only a small proportion of the concentrates yet prepared is the active erythropoietic material. Recent work by Campbell *et al.*[11,12] with sheep plasma has strengthened this conclusion. Their most active preparation is lower in sialic acid.

The cold, neutralized filtrate from heat-coagulated anæmic rabbit plasma[3] is mixed with 1·5 vol. of absolute ethanol and allowed to stand at 5° overnight. The inactive precipitate is removed by centrifugation in the cold and the supernatant mixed with an equal volume of absolute ethanol. After standing overnight, the precipitate is collected in the cold and dried in a desiccator. It is extracted with 20 parts of 0·01 *M* sodium acetate, *p*H 4·8, and the soluble portion (about 85 per cent) is dialysed against distilled water. The dialysand is lyophilized. 60–120 mgm. of a very soluble white powder are obtained from 1 litre of plasma. We will refer to this preparation as *LD*.

When *LD* is applied to a column of 'DEAE' cellulose (1 gm.) equilibrated in 0·01 *M* sodium acetate, *p*H 4·8, 15–30 per cent of protein emerges as a front, 10–20 per cent as another peak with 0·05 *M* sodium chloride as eluant. Both are erythropoietically inactive. Elution with 0·1 *M* sodium chloride yields a peak containing all the activity and 50–60 per cent of the protein. It again gives a single peak when rechromatographed on a similar column. Its properties are listed in Table 1.

A potent human urinary ultrafiltration residue[9], a gift of Dr. Van Dyke, was applied to a 'DEAE' column as above. 0·25 *M* sodium chloride had to be used for elution of the activity. Paper electrophoresis at *p*H 8·6 and 4·5 showed it to be similar to *LD* (mobility like an acid α_1 protein). Similar behaviour is shown by erythropoietin obtained from unboiled sheep plasma before its final purification[11,12].

Table 1

	Buffer	*p*H	Ionic strength	Mobility $\times 10^5$ cm.^{2} volt sec.
Moving boundary electrophoresis.	Sodium barbiturate	8 75	0·1	– 5 40
	Sodium acetate	4·25	0 1	– 2·92
	Glycine hydrochloride	2·34	0·1	+ 0·79
Isoelectric point		2·75		
E(1 per cent, 1 cm) 280 mμ		7·0		
Protein bound hexose (orcinol)		9 4%		
Hexosamine (Elson-Morgan, Rimington)		9 0%		
Sialic acid (resorcinol hydrochloride, Svennerholm (ref. 17) 15 min., 100° C)		13–14%		
Methyl pentose		0 66–0·80%		

Neuraminidase treatment. (*a*) 15 mgm. *LD* in 2·4 ml. 0·05 *M* calcium acetate, *p*H 6, were incubated with about 30,000 units of neuraminidase[8] at 35°. After 1 hr., 0·1 ml. of 1 *M* sodium citrate and 10 ml. water were added, and the mixture boiled for 5 min. It was erythropoietically inactive. No loss of activity was found in a control experiment without enzyme added. (*b*) In similar experiments the reaction was stopped by sodium citrate alone without boiling after 10 min. and 1 hr. respectively, and each reaction mixture was then dialysed against 10 vol. of water. In both cases practically all the sialic acid of *LD* was found in the dialysate, and both dialysates and dialysands were erythropoietically inactive. Peptide bond hydrolysis (increase in amino-nitrogen) was insignificant and probably did not occur.

A number of relatively mild substitution reactions were carried out with *LD* :

(1) *Iodination* (ref. 13). To 10 mgm. *LD* in 0·4 ml. 0·35 *M* sodium phosphate *p*H 7·6, increments of 0·1 *N* iodine were added at room temperature. 0·07–0·13 ml. were taken up in 3½ hr.

(2) *Acetylation* (ref. 14). To 10 mgm. *LD* in 0·2 ml. of a solution of 11·9 gm. sodium acetate in 20 ml. water (*p*H = 9·25) 0·03 ml. of acetic anhydride was added over a period of 1 hr. at 0°.

(3) *Formaldehyde* (ref. 15). 10 mgm. *LD* in 0·1 ml. 0·35 *M* sodium phosphate *p*H 7·6 and 0·01 ml. 40 per cent formaldehyde were allowed to stand for 20 hr. at 8°. Excess formaldehyde removed *in vacuo*.

(4) *Esterification* (ref. 16). To 10 mgm. *LD* in 0·4 ml. methanol at –15° 0·02 ml. of thionylchloride were added over 3 hr. Methanol was removed *in vacuo*.

All four of the above reaction mixtures were erythropoietically inactive. (Assayed by iron-59 uptake in fasted rats[3,18].)

The enzymatic loss of erythropoietic activity indicates that bound sialic acid is an essential part of the molecule. It remains to be seen whether the high sensitivity of the molecule to substitution reactions carried out under conditions too mild for hydrolysis is related only to the sialic acid moiety or other sugar or amino-acid constituents of the active mucoprotein.

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[1] Lowy, P. H., Keighley, G., Borsook, H., and Graybiel, A., *Nature*, 181, 1802 (1958).
[2] Lowy, P H., Keighley, G., and Borsook, H., *Proc. Soc. Exp. Biol. Med.*, 99, 668 (1958).
[3] Lowy, P. H., Keighley, G., Borsook, H , and Graybiel, A., *Blood*, 14, 262 (1959).
[4] Rambach, W. A., Alt, H. L., and Cooper, J. A. D., *Blood*, 12, 1101 (1957).
[5] Slaunwhite, jun., W. R., Mirand, A. E., and Prentice, T. C., *Proc. Soc. Exp. Biol. Med* , 96, 616 (1957).
[6] Rambach, W. A., Shaw, R. A., Cooper, J. A. D., and Alt, H. L., *Proc. Soc. Exp. Biol. Med.*, 99, 482 (1958).
[7] Boström, H., Roden, L., and Yamashina, I., *J. Biol. Chem.*, 230, 381 (1958).
[8] Ada, G. L., and French, E. L., *Nature*, 183, 1740 (1959).
[9] Garcia, J. F., and Van Dyke, D. C., *J. App. Physiol.*, 14, 233 (1959).
[10] Rambach, W. A., Cooper, J. A. D., and Alt, H. L., *Proc. Soc. Exp. Biol. Med.*, 98 602 (1958).
[11] Campbell, B. J., Schlueter, R. J., White, W. F., and Weber, G. F., Abstracts 66 C., 136th meeting Amer. Chem Soc., Atlantic City (1959).
[12] Goldwasser, E., and White, W. F., *Fed. Proc.*, 18, 236 (1959).
[13] Fraenkel-Conrat, J., and Fraenkel-Conrat, H., *Biochim. Biophys. Acta*, 5, 89 (1950).
[14] Fraenkel-Conrat, H., Bean, R. S., and Lineweaver, H., *J. Biol. Chem.*, 177, 385 (1949).
[15] Fraenkel-Conrat, H., and Olcott, H. S., *J. Amer. Chem. Soc.*, 70, 2673 (1948).
[16] Bello, J., *Biochim. Biophys. Acta*, 20, 426 (1956).
[17] Svennerholm, L., *Biochim. Biophys. Acta*, 24, 604 (1957).
[18] Fried, W., Plzak, L. F., Jacobson, L. O., and Goldwasser, E , *Proc. Soc. Exp. Biol. Med.*, 94, 237 (1957).

Co-factors in *Hevea brasiliensis* Latex Serum

IT has recently been shown[1] that the incorporation of mevalonic acid into rubber by an enzyme system in *Hevea* latex *in vitro* is not dependent on the addition of such co-factors as are known to be necessary for the incorporation of mevalonic acid into squalene. Such co-factors include adenosine triphosphate, cysteine and/or glutathione in the case of cell-free preparation of rat liver[2], although Amdur, Rilling and Bloch[3] found that whereas adenosine triphosphate was necessary with a soluble extract of bakers' yeast, no reducing agent requirement was demonstrated. Popjak and Gore show that under anaerobic conditions the reducing agent is not necessary but it is essential for the formation of squalene aerobically. Tchen[4] has demonstrated that the purified enzyme 'mevalonic acid-kinase' responsible for part of the pathway, namely, the phosphorylation of mevalonic acid, requires the presence of any of the four nucleoside triphosphates (including cytidine triphosphate). He shows also that the enzyme is inhibited by chloromercuribenzoate.

The present communication is to record the detection and assay of di- and tri-phosphates of adenosine and cytidine and of cysteine and reduced glutathione in samples of freeze-dried fresh latex sera obtained from Malaya. This material[5] would be expected to be equivalent to the serum from fresh latex 1–2 hr. after tapping, so far as soluble co-enzyme content is concerned. Details of these investigations will be published elsewhere; but it is thought of interest to record that such samples of normal latex sera examined contain approximately 0·7 micromole of glutathione and 0·4 micromole of cysteine per ml. and at least 0·0005 per cent nucleoside diphosphate in the form of adenosine diphosphate and cytidine diphosphate (predominantly as adenosine diphosphate), and almost the same amount of nucleoside triphosphate (predominantly as cytidine triphosphate).

It would be of interest to determine the influence of —SH inactivating agents on both the aerobic and anaerobic incubation of mevalonic acid labelled with carbon-14 in fresh latex to establish the similarity or otherwise of the general reductive mechanism in the two instances.

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[1] Park, C., and Bonner, J., *J. Biol. Chem* , 233, 340 (1958).
[2] Popjak, G., Gosselin, L., and Gore, I. Y., *Biochem. J.*, 70, 15*p* (1958).
[3] Amdur, B. G., Rilling, H., and Bloch, K., *J. Amer. Chem. Soc.*, 79, 2647 (1957).
[4] Tchen, T. T., *J. Biol. Chem.*, 233, 1100 (1958).
[5] McMullen, A. I., *Biochem. J.*, 72, 545 (1959).

Isolation of the Neurotoxic Component of the Venom of the Sea Snake, *Enhydrina schistosa*

THE sea snake, *Enhydrina schistosa*, is widely distributed in the coastal regions of the Far East and many fatalities from its bite are reported[1]. The *LD*50 of the venom for a wide variety of laboratory mammals is in the region of 50–100 μgm./kgm. body-weight. On this basis it is four times as toxic as the venom of cobra. Venom yields per bite of up to 55 mgm. of dried venom have been obtained from captive snakes. If humans are of equal susceptibility to laboratory animals, up to 15 *LD*50 for humans may be expected from one bite. At present there is no supply of antivenom available for treatment, and it is doubtful if the titres obtainable by even the best available preparative methods would be sufficient to allow the antivenom being administered in a life-saving dose. The separation of the neurotoxic component of this venom is important from the point of view of analysing its biochemical actions and enhancing its antigenic properties.

Carey and Wright[2] have shown that the whole venom consists of at least three antigenic components. The neurotoxic component is the most electropositive (*p*H 6·8 in agar gel) and will pass through a cellulose but not a nylon dialysis sac.

In the present investigation it has been found that after repeating the cellulose sac dialysis against large volumes of distilled water approximately 1 per cent of the initial toxicity remains inside the sac. This may be due to a different toxic component or to adsorption of traces of dialysable component on to non-dialysable material. However, it is unlikely to contribute significantly to the toxicity of the whole venom. Immunoelectrophoresis of the cellulose sac dialysate using antiserum to the whole venom shows only one main precipitation line. However, this has since been shown to consist of at least two antigenic components by varying the relative concentrations of venom and antiserum in the immunoelectrophoresis. When the dialysate was placed on a carboxy methyl cellulose column and eluted with *p*H 6·6 buffers of graded molarities from 0·06 to

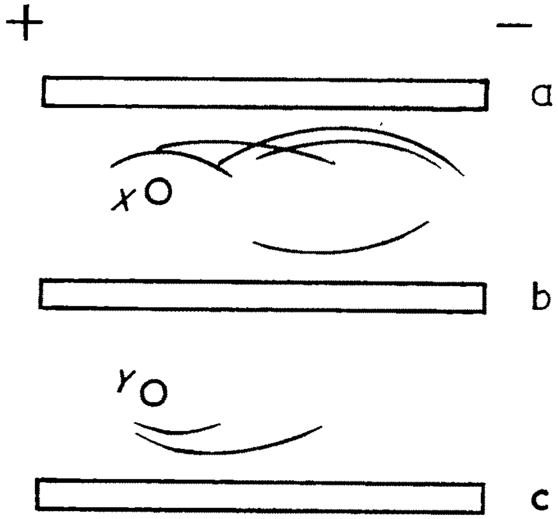

Fig. 1. A diagram of the immunoelectrophoretic patterns obtained with *Enhydrina schistosa* venom at *p*H 6 8 in phosphate buffer and electrophoresed for 3 hr. at 4 volts/cm. *a*, Antiserum to the whole venom in channel; *b*, antiserum to the toxic fraction in channel; *c*, antiserum to the whole venom in channel; *X*, whole venom at origin; *Y*, whole venom mixed with excess antiserum to the toxic fraction at origin

0·08 *M*, a further purification of the toxic component was obtained. Toxicity equivalent to 80–100 per cent of that of the initial sample was recovered in the eluant in one sharply defined zone. Immunoelectrophoresis of this sample using antiserum to the whole venom gave one precipitation line only over a range of relative concentrations of venom and antiserum. When whole venom is mixed with excess antiserum prepared against the toxic fraction only, the supernatant is non-toxic but still contains antigenic material as demonstrated by immunoelectrophoresis. The immunoelectrophoretic patterns of this supernatant material compared with that of the whole venom using antiserum to the whole and fractionated venom are shown in Fig. 1. An antiserum to this one fraction may therefore be as useful therapeutically as an antiserum to the whole venom. There was, however, no demonstrable advantage in using the purified toxic fraction as the antigen in the preparation of a protective serum.

Conflicting reports have appeared in the literature regarding the importance of a lecithinase in the toxicity of some venoms[3-5]. For demonstrating lecithinase activity we incubated the venom with egg yolk and then tested the solution for hæmolytic activity. After dialysis using a 'Cellophane' membrane, more than 90 per cent of the lecithinase activity of the whole venom could be retained inside the sac while more than 90 per cent of the initial toxicity passed through. This distinction between the two factors is further supported by the finding that the toxicity of the whole venom is more readily destroyed by heating than is its lecithinase activity.

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[1] Reid, H A., and Lim, K J., *Brit. Med J*, ii, 1260 (1957).
[2] Carey, J. E., and Wright, E. A., *Trans. Roy. Soc. Trop. Med. Hyg.* (in the press).
[3] Slotta, K. H., and Fraenkel Conrat, H., *Ber.*, 71, 1076 (1938).
[4] Fraenkel Conrat, H, and Singer, B., *Arch. Biochem. Biophys.*, 60, 64 (1956).
[5] Detrait, J., and Boquet, P., *C.R. Acad. Sci., Paris*, 246, 1107 (1958).

Transglucosidase Activity in *Acetobacter xylinum*

Cells of *Acetobacter xylinum* are known to produce cellulose. This synthesis has been demonstrated in resting fresh and lyophilized cells[1,2]. Enzymic synthesis of cellulose by preparations from *A. xylinum* has been shown with the aid of tracer techniques[3,4] and electronmicrography[5].

Glaser's experiments[4] with uridine diphosphoglucose labelled with carbon-14 in the glucose moiety demonstrate the incorporation of glucose into cellulose via uridine diphosphoglucose:

$$\text{uridine diphosphoglucose} + (\text{glucose})_n \longrightarrow \text{uridine diphosphate} + (\text{glucose})_{n+1}$$

No experimental evidence has been presented for the reverse reaction, that is, the formation of uridine diphosphoglucose from uridine diphosphate and soluble cellodextrins. If the reaction is reversible, cellodextrin molecules may serve both as donors and acceptors of glucose units; the donors thereby become shorter, while the acceptors increase in length. Most likely there is a certain probability for obtaining some alkali-insoluble chains of high molecular weight.

In the present experiments a net increase of alkali-insoluble carbohydrate material was found upon the addition of uridine diphosphate and soluble cellodextrins (the soluble cellodextrins were made by a modified procedure of the method given by Zechmeister *et al.*[6]) to a preparation from *A. xylinum*.

Pellicles of *A. xylinum* (N.C.I.B.8246) were freed from cellulose by the method of Hestrin and Schramm[2], the cells were ruptured, with careful *p*H control, with an ultrasonic disintegrator, and centrifuged to eliminate remaining cells and cell-debris. In some experiments the cell-free extract was used as such. Attempts were made to fractionate the extracts by collecting the precipitates at 19,000*g* and 35,000*g*, and resuspending the pellets in 0·1 *M tris*(hydroxymethyl)aminomethane buffer of *p*H 8. The reaction was carried out in centrifuge tubes kept in an evacuated container at 30° C. After 20 hr. NaOH was added to a final concentration of 1 per cent, and the tubes were heated in a boiling water-bath for

Table 1

Experiment	Additions	Alkali-insoluble material formed. As: Hexose (μgm)	Net increase (per cent)
1	None	180	—
	Uridine diphosphoglucose	200	11
	Uridine diphosphoglucose, cellodextrins	320	78
	Cellodextrins	220	22
2	None	8	—
	Uridine diphosphoglucose	12	50
	Uridine diphosphoglucose, cellodextrins	17	112
	Cellodextrins	14	75
3	None	70	—
	None	68	—
	Uridine diphosphate, cellodextrins	188	172
	Cellodextrins	118	71
	Uridine diphosphate, cellodextrins, cellobiose	184	167
	Cellodextrins, cellobiose	94	36
	Uridine diphosphate, cellobiose	56	—
	Cellodextrins, unincubated	64	

Each tube contained 1 ml. enzyme preparation of *p*H 8, plus additions as given, in the following amounts uridine diphosphoglucose, 8 5 μmoles; uridine diphosphate, 9·7 μmoles; cellobiose, 59 μmoles; cellodextrins, 15–20 mgm.; total volume per tube, 2 ml Different enzyme preparations were used in the three experiments. In 1, the pellet from 19,000*g*; in 2, the pellet from 35,000*g*; and in 3, the non-fractionated extract.

5 min. and then centrifuged. The precipitates were washed twice with water and dried. Hexose was determined by the anthrone method[7]. The results are presented in Table 1.

These results indicate the presence of transglucosidase activity in the preparation from *A. xylinum*. The insolubilization of soluble cellodextrins through the action of transglucosidase will from statistical reasons be expected to give low yields of high molecular weight compounds. Further work is in progress to purify to some extent the enzyme system in order to investigate the nature and specificity of co-factors in the reaction.

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[1] Hestrin, S., Aschner, M., and Mager, J., *Nature*, 159, 64 (1947).
[2] Hestrin, S., and Schramm, M., *Biochem. J.*, 58, 345 (1954).
[3] Greathouse, G. A., *J. Amer. Chem. Soc.*, 79, 4503 (1957).
[4] Glaser, L., *J. Biol. Chem.*, 232, 627 (1958).
[5] Colvin, J. R., *Arch. Biochem. Biophys.*, 70, 294 (1957).
[6] Zechmeister, L., and Toth, G., *Ber.*, 64, 854 (1931).
[7] Viles, jun., F. J., and Silverman, L., *Anal. Chem.*, 21, 950 (1949).

Preparation of Crystalline Cytochrome$_{553}$ from *Porphyra tenera*

PARTIAL purification of a cytochrome component from *Porphyra tenera*, a component which has absorption peaks similar to that of cytochrome *c* or c_1 was first reported by Yakushiji[1]. Recently, Katoh and Takamiya (ref. 2 and Katoh, S., and Takamiya, A., personal communication) succeeded in obtaining a crystalline preparation of this component by means of diethylaminoethyl-cellulose column chromatography and reported the redox-potential of this and other components in marine algae. We also have crystallized the cytochrome by means of a cation-exchanger at *p*H 5·0. Since the diethylaminoethyl-cellulose column procedure is possible at *p*H 7·0, this procedure may be better; our method can be used to purify not only basic enzyme proteins, but also those that are stable in acidic *p*H.

About 700 gm. of the algae dried by exposure to the Sun was suspended in 12 l. of water and kept for 3 days at 15°. The suspension was squeezed through a cheese-cloth and about 11 l. of reddish-violet extract obtained. To this extract solid ammonium sulphate was added to obtain a concentration of 50 per cent saturation and the resulting precipitate, which contains almost all phycobilin, was removed from the extract by filtration. To recover the cytochrome from the precipitate, the precipitate was dissolved in 1 l. of 0·05 *M* disodium hydrogen phosphate and the above fractionation was repeated with ammonium sulphate. Both filtrates were combined and the cytochrome precipitated by further addition of ammonium sulphate to raise the concentration to 100 per cent saturation. The precipitate, collected by filtration with the aid of 'Celite', was dissolved in about 500 ml. of 0·05 *M* disodium hydrogen phosphate and 1 per cent of rivanol added drop by drop to the solution until fibrous substances no longer formed. These substances were removed by filtration and the filtrate fractionated with ammonium sulphate to obtain a fraction between 80 and 100 per cent saturation. By repeating this fractionation twice, an excess of rivanol was removed. The precipitate was dissolved in 50 ml. water and dialysed against 2 l. of 0·02 *M* sodium acetate buffer of *p*H 5·0 overnight at 5°. After the dialysis, the solution was centrifuged to remove dark brown precipitates which appeared during the dialysis. The clear supernatant fluid was then charged on the top of column (1 × 50 cm.) of a cation-exchanger ('Amberlite *CG*–50', type II) buffered at *p*H 5·0 with the same buffer. The column adsorbed the cytochrome and was washed with 2 l. of the same buffer and then 2 l. of 0·1 *M* sodium acetate buffer, *p*H 5·0. These washings eluted some of the impurities and modified cytochrome. The adsorbed cytochrome was then eluted with 0·1 *M* sodium acetate buffer of *p*H 5·8. About 1 l. of eluate contained cytochrome which was concentrated by treatment with polyethyleneglycol[3]. This chromatography was carried out in the oxidized state, though chromatography is also possible in the reduced state at *p*H 5·0. In the latter case, however, an affinity of the cytochrome for resin is less than that of the oxidized state, so that the elution process was performed with 0·02 *M* buffer at *p*H 5·0 and it is necessary to repeat the chromatography twice in order sufficiently to remove the impurities. To the eluate solid ammonium sulphate was added to precipitate the cytochrome completely. The cytochrome was collected by centrifugation and dissolved in a minimum amount of water. Insoluble material is removed by centrifugation, and solid ammonium sulphate was added to the supernatant fluid by a small glass rod in several portions until small turbidities appeared. The solution was kept in a tap-water bath and the turbidity slowly increased. Within 2–3 hr., the crystals began to appear and almost all the cytochrome crystallized as needles within 1 day. The crystals were collected by centrifugation, washed with 90 per cent saturated ammonium sulphate solution, dissolved in a minimum amount of water, and then recrystallized as above.

Fig. 1 shows the absorption spectrum of a once-recrystallized preparation dissolved in 0·1 *M* phosphate buffer of *p*H 7·4. The α-band of the cytochrome shows its peak at 553 mμ and a shoulder at about

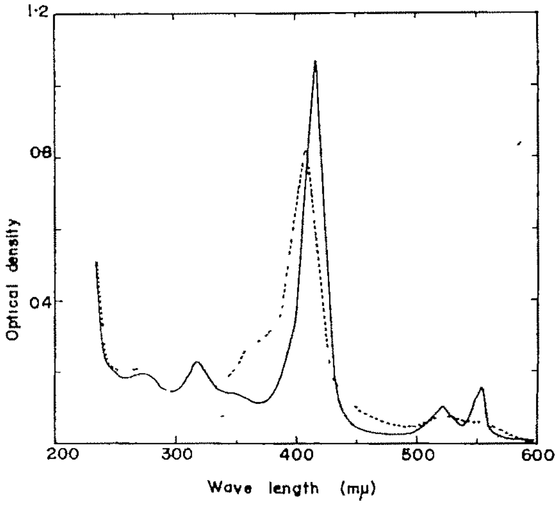

Fig. 1 Absorption spectrum of cytochrome$_{553}$ from *Porphyra tenera*. The crystals were dissolved in 0·1 *M* phosphate buffer of *p*H 7·4 and dialysed against the same buffer. Measurements are carried out by a Cary spectrophotometer Model 14 by using a 1-cm. light-path cuvette. The dotted line shows the oxidized form and the solid line the reduced form after the addition of sodium borohydride

550 mμ. However, other peaks are similar to that of beef cytochrome c or c_1. The absorption peaks of pyridine hæmochromogen at a high pH are similar to that of cytochrome c and the α-peak has no longer a shoulder. The hæm cannot be split from the protein moiety by the usual acid–acetone method (1 ml. 20 per cent hydrochloric + 100 ml. acetone). From these results it appears that the hæm of the cytochrome belongs to hæm c. The reduced cytochrome is not autoxidizable and is oxidized by cytochrome a_2 purified from *Pseudomonas aeruginosa*[4], but not by mammalian cytochrome a[5].

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[1] Yakushiji, E., *Acta Phytochim.*, 8, 325 (1935).
[2] Katoh, S., *J. Biochem. (Japan)*, 46, 629 (1959).
[3] Kohn, J., *Nature*, 183, 1055 (1959).
[4] Horio, T., Higashi, T., Matsubara, H., Kusai, K., Nakai, M., and Okunuki, K., *Biochim. Biophys. Acta*, 29, 297 (1958).
[5] Okunuki, K., Sekuzu, I., Yonetani, T., and Takemori, S., *J. Biochem. (Japan)*, 45, 847 (1958).

ANIMAL PHYSIOLOGY

Withdrawal of Inhibition of Mitosis as a Mechanism Influencing Normal and Pathological Growth

EXPERIMENTAL evidence has recently been given[1] of the likelihood that wound-healing occurs not from the stimulation of wound hormones directly, as has been thought for many years, but by the withdrawal of inhibiting influences as a result of cell damage.

Several experiments performed in this laboratory using regenerating liver of rats and guinea pigs have demonstrated that corticosteroids act as inhibitors to growth, and circumstances in which these substances are withdrawn or reduced have simulated a 'growth hormone effect'. From these observations a mechanism has been proposed which could account for some features of rapid tumour growth, and attention has been directed to the similarity of conditions in wound healing[2].

The histological appearances and mitotic index have been found to be similar in three different systems which have in common a reduced corticosteroid to growth hormone ratio.

In the first system, Cater, Holmes and Mee[3] showed that injections of pure growth hormone to intact rats caused an increased mitotic rate and synthesis of deoxyribonucleic acid. The mitotic index was approximately double that found in normal rat regenerating liver. The influence of pituitary hormones and cortisone on liver regeneration rate in hypophysectomized rats was then investigated[4]. Cortisone caused complete inhibition of mitosis, which was overcome proportionately as the dose of growth hormone was increased. Growth hormone alone to hypophysectomized rats did not increase the rate of mitosis after partial hepatectomy.

The second system used was the regenerating liver of the adrenalectomized rat in which the rate of mitosis and histological appearances are identical with those of intact rats given growth hormone. A large number of such rats were examined and a study made of the influence of time between adrenalectomy and partial hepatectomy (unpublished work). During the first 48 hr. no inhibiting influence is present and an approximate doubling of normal regeneration rate is seen but after 7 days a return to normal mitotic index occurs.

The third system involved liver regeneration in rats bearing Walker 256 carcinomas. Observations were made of resting liver mitoses, of the mitotic rate 29 hr. after partial hepatectomy, of adrenal weights and of liver glycogen as an indication of corticosteroid output at that time. Two groups were compared, rats bearing large fast-growing tumours and those bearing small slow-growing tumours. It was found that resting liver mitosis rates averaged seven times greater in tumour-bearing rats than in normal rats. Regenerating liver from the fast-growing tumour-bearing rats had a very high mitotic index, not significantly different from that found in adrenalectomized rats. Regenerating liver of slow-growing or arrested tumour-bearing rats had a mitotic index equal to or less than that of normal rat regenerating liver. Liver glycogen in the two groups was significantly different, the larger fast-growing tumour rats tending to have less liver glycogen than liver from a normal rat, while the slow-growing tumour rat deposited about twice the liver glycogen of a normal rat. These glycogen estimations were not regularly consistent but are believed to indicate trends in circulating corticosteroid values.

Taken together, the findings from tumour-bearing rats suggest lower corticosteroid levels in those rats with the fastest-growing tumours. The liver mitosis rates and histological appearances are similar to those found in adrenalectomized rat regenerating liver and the growth hormone-treated rat. These are 'growth hormone like' appearances due in the first two sytems to reduction in the ratio of corticosteroids to growth hormone. In the third system of tumour-bearing rats either a reduction of circulating corticosteroids or some reduction of their effectiveness would account for the similar mitotic index and histological appearances.

Some evidence is being obtained from experiments not yet completed that corticosteroids can be rendered ineffective with regard to their influence on mitosis. It is known from system two mentioned above that removal of corticosteroids from the rat allows an approximate doubling of the normal rate of mitosis in regenerating liver. From experiments of workers[5,6] in the field of ascorbic acid relationship to corticosteroid synthesis and as a direct result of an experiment demonstrating corticosteroid influence on the radiation effect[7], it seemed possible that certain redox chains depending for their integrity on the presence of ascorbic acid were permissive in allowing corticosteroid activity. It seemed that a 'functional adrenalectomy' might be obtained in the guinea pig by deprivation of ascorbic acid so that partial hepatectomy in this animal (unreported work) might reveal a doubling of mitosis-rate over that of normal guinea pig regenerating liver, as has been found in the adrenalectomized rat. The difficulties in this procedure are being overcome and will form a full report as soon as statistically satisfactory results are obtained. To date 9 normal guinea pigs have survived partial hepatectomy and show a mean regeneration rate of 24 mitoses per 1,000 cells. Mortality among scorbutic guinea pigs is high but of 30 animals 3 have survived with a mean mitotic index of 50. This is a positive result which is unlikely to occur under any other conditions and each individual result should have meaning. To support this, 10 other animals have survived which were believed to have been fully scorbutic but which showed no mitoses in the liver after the same regeneration time. This is consistent with the idea of a low threshold-level of ascorbic acid, and until this is reached, adequate though small amounts of ascorbic acid permit corticosteroid inhibition of mitoses. Very high levels of circulating corticosteroids are known to occur in scorbutic animals[8] and these undoubtedly inhibit all mitoses.

If withdrawal of corticosteroid influence is a likely means of instigation of growth processes, some special mechanism must occur in wound-healing to overcome locally the influence of the raised corticosteroid circulating levels which are inimical to the processes of inflammation and repair. It is suggested that damaged cells may secrete or produce an oxidizing agent in minute amounts which exerts an influence only in the area of damage. Thus neither ascorbic acid nor corticosteroid levels in the circulation need be depleted and may not be found changed on investigation. It is known that following X-irradiation in animals excretion of dehydro-ascorbic and di-ketogulonic acids is increased[9].

These studies have had as an object the mechanism of failure of control of tumour cell growth and it seems likely that tumours act as damaged cells. In addition to their own point of damage which shows itself as, among other things, a failure to respond to circulating corticosteroids[10], it is likely that a spreading influence, at first confined to the immediate cell boundary, but later in an attenuated form perfusing the circulation, renders the host more liable to establishment of metastases and to multiple tumours. Some very recent evidence[11] supporting this concept is of the growth of dormant injected tumour cells in the liver after mere handling of this organ. The fact that an injected tumour-cell population needs to be of a minimum size for survival suggests some ability to influence the host which may not be only of an antigenic nature.

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[1] Bullough, W. S., Conference on Wound Healing, Royal College of Surgeons of England (1959).
[2] Hemingway, J. T., Ph.D. thesis submitted to Univ. of Cambridge (1959).
[3] Cater, D. B., Holmes, B. E., and Mee, L. K., *Biochem. J*, 66, 482 (1957).
[4] Hemingway, J. T., and Cater, D. B., *Nature*, 181, 1065 (1958).
[5] Banergee, S., and Debb, C., *Endocrinol.*, 51, 572 (1952).
[6] Bacchus, H., and Altzuler, N., *Endocrinol.*, 51, 1, 578 (1952).
[7] Hemingway, J. T., *Experientia*, 15, 189 (1959).
[8] Burnstein, S., Dorfman, R., and Nadel, E. M., *J. Biol. Chem.*, 213, 597 (1955).
[9] Monier, M. M., and Weiss, R. J., *P.S.E.B.M.*, 81, 59 (1952).
[10] Green, H. N., and Savigear, M., *Brit. Med. J.*, 1, 498 (1951).
[11] Fisher, E. R., and Fisher, B., *Science*, 130, 918 (1959).

Effect of Chlorpromazine on Tissue Metabolism *in vitro*

IT has been pointed out that central inhibition is connected with an increase of acetylcholine content in brain tissue. On the other hand it has also been shown that some central depressants reduce the oxygen uptake of normal respiring brain tissue *in vitro*[1] or in brain slices which were stimulated electrically[2].

In previous experiments we have seen that chlorpromazine as a tranquillizing drug can also enhance the acetylcholine content in the brain of rats[3]. Further results have confirmed the inhibitory effect of chlorpromazine (even in low doses of 1–10 µgm./ml.) on the rate of oxygen uptake in brain homogenates metabolizing glucose as substrate. However, chlorpromazine did not inhibit uptake of oxygen in the presence of succinate[4].

Studying these relationships we have investigated the direct influence of acetylcholine on the respiratory activity of brain homogenates (albino rats) *in vitro*. The oxygen uptake was followed by the direct Warburg technique (buffered Krebs–Ringer phosphate solution, pure oxygen, 37° C.). The brain tissue was homogenized in the presence of eserine. Results are given in Table 1. Eserine alone did not change the respiration significantly. The lack of some changes after acetylcholine is in agreement with the findings of McIlwain[5] obtained in brain slices of the rat. Similar negative results were observed with the oxygen uptake of hemi-diaphragm slices incubated with acetylcholine.

The inhibitory effect of chlorpromazine on the endogenous respiration (no substrate added) of brain homogenates was reached at relatively high concentration of this substance only. This effect was deepened when ammonium chloride was added simultaneously with chlorpromazine (Table 2).

Contrary to the results of Abood[6] obtained in a mitochondrial preparation of the brain, we could not confirm the inhibitory effect of chlorpromazine on cytochrome oxidase activity in brain homogenates. Enzyme activity was followed by oxidation of 5 m*M* *p*-phenylenediamine during the first hour after addition of chlorpromazine to the brain homogenate. In controls the mean value with standard error of 6 exp. was 143·6 ± 4·7 (µl. oxygen)/100 mgm. wet

Table 1. OXYGEN UPTAKE OF BRAIN HOMOGENATES AND DIAPHRAGM SLICES AFTER ESERINE AND ACETYLCHOLINE. MEDIUM: BUFFERED KREBS–RINGER SOLUTION WITH 0·2 PER CENT GLUCOSE. CHANGES IN PERCENTAGE ARE NOT SIGNIFICANT

No. of experiment	Tissue	Treatment	µl. O_2/100 mgm. wet w./hr.		Changes (per cent)
			Mean	*S.E.*	
6	Brain homogenate	Control	112·77	± 1·63	
6		Eserine salicyl. (83 µgm./ml.)	107·32	± 2·93	– 4 85
6	Brain homogenate	Eserine salicyl. (83 µgm./ml.)	107 32	± 2·93	
		Eserine salicyl. (83 µgm./ml.) + acetylcholine/ml.:			
5		10 µgm.	106 91	± 4·07	– 0·39
5		100 µgm.	110 85	± 4·77	+ 3 28
5		300 µgm.	109·13	± 3·44	+ 1·68
6		833 µgm.	109·29	± 3·29	+ 1·88
9	Hemidiaphragm slices	Eserine salicyl. (83 µgm./ml.)	96 48	± 4 45	
9		Eserine salicyl. (83 µgm./ml.) + acetylcholine (100 µgm./ml.)	99·66	± 5·52	+ 3·3

Table 2. INHIBITION OF THE ENDOGENOUS RESPIRATION AFTER ADDITION OF CHLORPROMAZINE AND AMMONIUM CHLORIDE IN BRAIN HOMOGENATES. MEDIUM: KREBS–RINGER SOLUTION (NO SUBSTRATE ADDED). THE INHIBITION IS SIGNIFICANT

No. of experiment	Treatment	µl. O_2/100 mgm. wet w./hr.		Inhibition (per cent)
		Mean	*S.E.*	
9	Control	82·72	± 1·98	
6	Chlorpromazine (200 µgm./ml.)	62·73	± 3·08	− 24·17
8	NH_4^+ (60 m*M*)	64·74	± 2·87	− 21·8
9	NH_4^+ (60 m*M*) + chlorpromazine (200 µgm./ml.)	51·87	± 3·9	− 37·3

weight (hr.) and after addition of 200 µgm. chlorpromazine/ml. the oxygen uptake was 143·2 ± 3·5.

We have reviewed the specificity of the action of chlorpromazine on cerebral tissue. It did not influence the oxygen uptake of rat's hemi-diaphragm slices metabolizing glucose as substrate. In control animals the mean value with standard deviation of 7 exp. was 95·5 ± 13·4 (µl. oxygen)/100 mgm. wet weight (hr.) and after 10 µgm. chlorpromazine/ml.—94·7 ± 10·3. In 6 exp. after addition of 200 µgm. chlorpromazine/ml. the oxygen uptake showed the value 94·4 ± 11·3.

It can be assumed that chlorpromazine produces an impairment in energy metabolism of the brain tissue such as depletion of oxygen uptake and inhibition of oxidative phosphorylation[7]. On the other hand chlorpromazine exerts no significant changes in cholinesterase activity of the brain as reported by Decsi[8] and it does not affect the synthesis of acetylcholine studied in brain homogenates of the rat[9]. These negative results favour the view that the increase of acetylcholine content following chlorpromazine is due to the depression of central activity without direct relation of chlorpromazine to the acetylcholine metabolism of the brain.

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[1] Jowett, M., *J. Physiol.*, 92, 322 (1938).

[2] McIlwain, H., *Biochem. J.*, 53, 403 (1953).

[3] Zelený, A., Lindaur, V., and Kozák, J., *Arch. Exp. Path. Pharmakol.*, 231, 593 (1957).

[4] Zelený, A., and Kozák, J., International Symposium: Pharmacology of the Nervous Excitability, Liblice, Č.S.R. (1959).

[5] McIlwain, H., *Brit. J. Pharmacol.*, 6, 531 (1951).

[6] Abood, L. G., and Romanchek, L., *Ann. N.Y. Acad. Sci.*, 66, 812 (1957).

[7] Decsi, L., *Arzneim.-Forsch.*, 8, 348, 352 (1958).

[8] Decsi, L., *Acta Physiol. Hung.*, 10, 387 (1956).

[9] Vlk, J., *Cs. fysiol.*, 7, 576 (1958).

Action of Thyroxine on the Rate of Oxygen Consumption of the Lung-Fish (*Protopterus*)

THE part that endocrines play in the control of metabolic rate of fishes has been investigated by numerous authors. The results concerning the action of thyroxine are contradictory[1–5]. This is mainly due to two essential factors: (1) feeding; (2) activity during measurements[6]. The current investigation was initiated to determine the effect of thyroxine upon the rate of oxygen consumption of *Protopterus*. These fish do not present the above-mentioned difficulties.

Two young individuals were captured from Tienaba (in the vicinity of Dakar), in February (during the aquatic phase), and were maintained in water until the beginning of the experiments in April. Each fish served as its own control, and thus individual variations were eliminated. The first presented external gills, the second only vestigial ones. The total wet weight of the fish was 27·5 and 46 gm. respectively. (The initial weight of the animals before the experiments was the only one taken into consideration.)

The rate of oxygen consumption was measured by means of Haldane's blood gas volumeter, slightly modified for this purpose. The reaction bulb was replaced by a respiratory chamber, inside which was introduced a small quantity of water, the animal and a small tube containing a carbon dioxide absorbent. This apparatus permitted us to measure the volume of oxygen absorbed by the animal to the nearest 0·01 c.c.

The normal rate of oxygen consumption of each individual was followed during the three days preceding the thyroxine injection and several readings taken. Each fish received a single intra-muscular injection of approximately 0·5 c.c. of thyroxine (Roche), in the dorsal muscles.

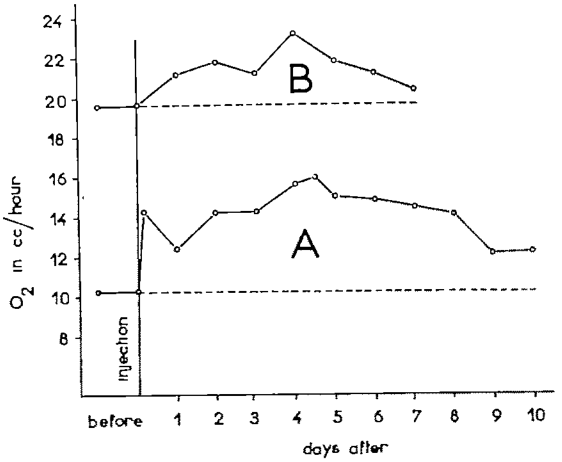

Fig. 1. Action of thyroxine on the rate of oxygen consumption of *Protopterus*

In both cases there was a remarkable increase in the rate of oxygen consumption, one day after injection (Fig. 1). The respiratory rate was maximal about the fourth day, then it dropped gradually during the following days in order to reach approximately its initial level.

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[1] Matty, A. J., *J. Marine Biol. Assoc. (U.K.)*, 33, 689 (1954).

[2] Drexler, E., and Issekutz, B., *Naunyn-Schmiedelberg's Arch. Exp. Pathol. Pharmakol.*, 177, 435 (1935).

[3] Etkin, W., Root, R., and Mogskin, B. P., *Physiol. Zool.*, 13, 415 (1940).

[4] Smith, D. C., and Matthews, S. A., *Amer. J. Physiol.*, 153, 215 (1948).

[5] Muller, J., *Z. vergleich. Physiol.*, 35, 1 (1953).

[6] Spoor, W. A., *Biol. Bull.*, 91, 312 (1946).

Presence of 5-Hydroxytryptamine after Raunescine in Blood Plasma *In vivo*

5-Hydroxytryptamine is localized in the platelets of the blood, minimal amounts being found in platelet-free plasma[1–3]. *Rauwolfia* alkaloids are known to lower the content of 5-hydroxytryptamine in various tissues including blood both *in vivo* and *in vitro*[4,5]. Whether 5-hydroxytryptamine is released by *Rauwolfia* alkaloids from the cellular binding sites as such to the cytoplasma is still not clear. It has been stated that the incubation of rabbit platelet suspensions with reserpine in an atmosphere of nitrogen increases the amount of 5-hydroxytryptamine in plasma[5].

We have investigated whether 'free' 5-hydroxytryptamine can be found in plasma after a *Rauwolfia* alkaloid. As in the earlier experiments[6], which originally directed our attention to this problem, we have used raunescine, which is an alkaloid chemically and pharmacologically closely related to reserpine[7].

3 ml. of blood was collected by using a polyethylene cannula from the abdominal aorta of male rats (160–180 gm.) anæsthetized with ether. Each time one of the rats served as a control and the three others that received 5 mgm./kgm. of raunescine intraperitoneally were bled 1, 3 and 6 hr. after the injection. The blood was mixed with 1/10 vol. of 1 per cent disodium ethylenediamine tetraacetate and 30–50 int. units of heparin/ml. After separation of plasma and platelets[8] the platelet-poor plasma was extracted by 19 vol. of acetone[9] and the packed platelets, after suspension in acid saline, by 10 vol. of acetone. The samples were kept at around 0° C. during the whole pre-extraction period. The biological assay of 5-hydroxytryptamine was made by using the rat stomach preparation[10]. For collecting the blood from the rabbits a catheter was inserted into a cervical vein under light ether anæsthesia, from which the animals were allowed to recover before the withdrawal of blood. In half of the experiments the animals were pre-treated with 100 mgm. (as base)/kgm. of iproniazid phosphate subcutaneously about 16 hr. before raunescine.

The results from rats are presented in Fig. 1. There was a gradual fall in the platelet 5-hydroxytryptamine, and after 6 hr. about 10 per cent was left in the non-pretreated rats and 14 per cent in those receiving iproniazid. Platelets were counted in some tests and they had about 1 µgm. of 5-hydroxytryptamine/10^9 cells on both series. Raunescine did not change the number of them. In the plasma there was a rise in both groups when analysed 1 hr. after raunescine administration. The increase was more than 10-fold in the pre-treated rats, being less marked in the others. The return to normal was also faster in the animals that received raunescine only.

Essentially similar but less consistent results were obtained by using rabbits. The increase of plasma 5-hydroxytryptamine was not as marked as in rats and it was negligible without iproniazid pre-treatment.

The 5-hydroxytryptamine in plasma probably comes from the platelets; but the possible release from other tissues cannot be excluded. The highest amounts of amine in the plasma were found at the time of the most marked release from the platelets. It has been reported that human serum oxidizes 5-hydroxytryptamine[11]. This activity could be inhibited by iproniazid, ceruloplasmin being suspected as the enzyme involved. Therefore the effect of iproniazid in our experiments may not be due to the inhibition of monoamine oxidase. It is interesting to note that the symptoms produced by raunescine become fully developed at a time when the plasma 5-hydroxytryptamine has returned to normal.

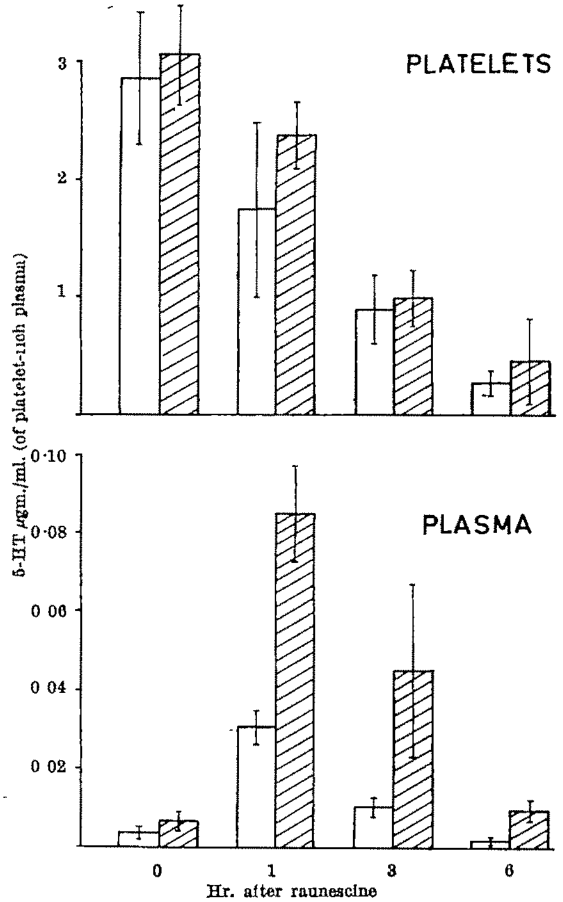

Fig. 1. The amounts of 5-hydroxytryptamine in platelets and platelet-free plasma of rats after raunescine. Each column represents the results from four experiments. Open columns, no pre-treatment; shaded columns, iproniazid pre-treatment; vertical lines, standard deviation

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[1] Zucker, M. B., Friedman, B. K., and Rapport, M. M., *Proc. Soc. Exp. Biol. and Med.*, 85, 282 (1954).

[2] Humphrey, J. H., and Jaques, R., *J. Physiol.*, 124, 305 (1954).

[3] Udenfriend, S., and Weissbach, H., *Fed. Proc.*, 13, 412 (1954).

[4] Shore, P. A., Pletscher, A., Tomich, E. G., Kuntzman, R., and Brodie, B. B., *J. Pharmacol.*, 117, 232 (1956).

[5] Carlsson, A., Shore, P. A., and Brodie, B. B., *J. Pharmacol.*, 120, 334 (1957).

[6] Paasonen, M. K., and Kärki, N. T., *Brit. J. Pharmacol.*, 14, 164 (1959).

[7] Paasonen, M. K., and Dews, P. B., *Brit. J. Pharmacol.*, 13, 84 (1958).

[8] Hardisty, R. M., and Stacey, R. S., *J. Physiol.*, 130, 711 (1955).

[9] Amin, A. H., Crawford, T. B. B., and Gaddum, J. H., *J. Physiol.*, 126, 596 (1954).

[10] Vane, J. R., *Brit. J. Pharmacol.*, 12, 344 (1957).

[11] Martin, G., Eriksen, N., and Benditt, E. P., *Fed. Proc.*, 17, 477 (1958).

Adrenocortical Function after Prolonged Salt Restriction in Hypertension

It has been suggested that the high salt intake current in most modern societies is beneficial by virtue of a presumptive stimulating action on the adrenal cortex[1]; it also has been reported[2] that *in vitro* production of corticosterone by the isolated adrenals of sodium-depleted rats declined progressively as a function of time, from which it has been inferred that limitation of salt intake might impair adrenocortical function resulting in an organism less responsive to stress. By contrast, there is considerable evidence implicating sodium in the hypertensive process both of man and animals[3]; furthermore, some results suggest that a high salt intake may be a causal factor in the etiology of human 'essential' hypertension—the second commonest cause of heart disease in Western society[4]. According to the latter view, a high salt diet would not be beneficial and might be frankly harmful.

It has been clearly established by many studies that under normal conditions metabolic balance can be maintained easily on daily salt intakes of 0·5–1·0 gm.—or approximately one-twentieth the amount commonly eaten by the average Western adult[4c]. A number of adrenocortical function studies made on man after short-term salt restriction have shown normal corticoid production with variable evidence of increased aldosterone production (for example, ref. 5). The long-term effects of salt restriction on adrenocortical function have not been studied but have assumed increasing importance with the widespread use of low salt diets in medicine: it is this aspect with which we are concerned here, brief mention of which was made in an earlier communication[4b]. A unique opportunity to make such observations was provided by 3 ambulatory subjects maintained on accurately measured diets, which contained only 100–150 mgm. of sodium (250–375 mgm. as sodium chloride) daily, for periods of from 1 to almost 5 years. We are aware of no natural diet which is so restricted in sodium as that used in the present work, and no suggestion is intended that comparable levels are necessary or advisable for the population generally: the present regimen for these patients was a deliberately severe test of the adaptive capacities of the adult organism carried out under careful supervision. The subjects included a 69-year old white widow and 2 white bachelors of 49 and 51 respectively (ages on admission). All had classical uncomplicated essential hypertension; they were ambulatory and went for long walks on the laboratory grounds ordinarily at least once daily. 24-hr. urine collections were made continuously and at no time was there evidence of extra salt intake. Numerous studies carried out on these individuals have been omitted unless clearly relevant to the current discussion. It is pertinent to note that on admission the 49-year old male was found to have an eosinophilia of 4–15 per cent which was not explained and which persisted in the hospital.

The observations reported here, unless otherwise indicated, were made after 58, 32 and 20 months on low salt regimen by the 69-year old woman, and the 49- and 51-year old men, respectively. Physically, in terms of disease entities or syndromes known to be related to the adrenal cortex, these subjects presented no evidence of either hypo- or hyper-corticism. All three had some personality peculiarities on admission which did not change with salt restriction: they appeared normal and acted normally and reported that they felt normal. The following laboratory studies, some of which may be abnormal with various states of adrenocortical dysfunction, were normal and essentially unchanged from the results obtained prior to salt restriction: plasma protein and specific gravity; hæmoglobin, hæmatocrit, white blood count and differential; urea clearance and blood urea nitrogen; plasma sodium, potassium, chloride and carbon dioxide; total exchangeable sodium as measured with sodium-24. Blood sugars remained normal both after eating and after a 24-hr. fast; intravenous glucose tolerance tests showed normal curves. The response to intravenous adrenocorticotrophic hormone[6] is summarized in Table 1. A normal fall in total circulating eosinophils occurred following the infusion of adrenocorticotrophic hormone, to 10 per

Table 1. 48-hr. Adrenocorticotrophic Hormone Test for Adrenocortical Reserve Function

Day	♀ 69				♂ 49				♂ 51			
	1	2	3	4	1	2	3	4	1	2	3	4
	(Control)		(Test)		(Control)		(Test)		(Control)		(Test)	
Eosinophils—mm.^{3} (normal: 100–250)	63	69	0	6	460	413	25	38	181	166	19	3
Urinary: 17-ketosteroids, mgm./24 hr. (normal: ♀ 5–15 ♂ 10–20)	9 7	10 0	9 0	15·1	10 0	8 2	14 5	28 9	13 0	14 5	21·1	23 8
Urinary: 17-hydroxy-corticoids, mgm./24 hr. (normal: ♀ + ♂ 4–11)	6·8	7 1	24 1	33 5	7·6	7·2	35·3	48·9	9·0	7·1	37·6	49 9
Months off sodium chloride	55				18				12			

Adrenocortical response to 25 units of adrenocorticotrophic hormone infused intravenously on both days 3 and 4.

Table 2. Urinary Steroid Excretion

	Normal range (per day)	♀ 69		♂ 49		♂ 51	
		Day 1	Day 2	Day 1	Day 2	Day 1	Day 2
Aldosterone	1–10 μgm.	Lost—laboratory accident	12	4	2	30	39
Cortisone	10–114 μgm.	175	163	10	13	62	64
Hydrocortisone	34–262 μgm.	309	412	(not done)		92	113
Months off sodium chloride		56		19		19	

cent or less of control values. The urinary excretion of both 17-ketosteroids and 17-hydroxycorticoids during the control days was within normal range and the subsequent response to the infusion of adrenocorticotrophic hormone indicated a normal adrenocortical reserve. Maintenance of sodium chloride balance during this period of severe restriction is *a priori* evidence of adequate production of salt-retaining hormones although no necessary indication of corticoid production. The daily urinary excretion of aldosterone, cortisone, and hydrocortisone[7] is summarized in Table 2. The significance of the slightly increased excretion of cortisone and hydrocortisone by the woman is doubtful in view of the wide range of normal, and in association with the normal values shown in Table 1 as well as the absence of other biochemical or clinical evidence of increased corticoid production. The aldosterone excretion for the 51-year old male appears elevated to levels reported for some studies[5] after short periods of salt restriction: the other two patients did not show this effect. From the results in Tables 1 and 2 we have concluded that prolonged salt restriction is associated with normal adrenocortical steroid production. We are aware that urinary excretion need not necessarily reflect adrenocortical production of steroids, and these results must be interpreted with this restriction. It is proper to ask whether the results reported here would hold in non-hypertensive individuals. We have subjected non-hypertensive persons to equal salt limitation for periods up to 3 months with similar findings, so it is unlikely that this capacity is peculiar to the individual with hypertension. Furthermore, during the past 12 years in an experience with more than 100 other individuals studied on this same regimen for periods as long as 1 year we have never found evidence of adrenocortical dysfunction.

From these studies, we cannot refute the suggestion that a high salt diet is beneficial but the evidence presented suggests that a prolonged and drastic low salt diet is compatible with normal adrenocortical function.

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[1] Kaunitz, H., *Nature*, 178, 1141 (1956).

[2] *a*, Eisenstein, A. B., and Hartroft, P. M., *Endocrinol.*, 60, 634 (1957). *b*, Hartroft, P. M., and Eisenstein, A. B., *ibid.*, 60, 641 (1957)

[3] Meneely, G. R., *Amer. J. Med.*, 16, 1 (1954).

[4] *a*, Dahl, L. K., and Love, R. A., *J. Amer. Med. Assoc.*, 164, 397 (1957). *b*, Dahl, L. K., *Nature*, 181, 989 (1958). *c*, Dahl, L. K., *New Eng. J. Med.*, 258, 1152, 1205 (1958).

[5] *a*, Crabbé, J., Reddy, W. J., Ross, E. J., and Thorn, G. W., *J. Clin. Endocrinol. and Metab.*, 18, 1147 (1958). *b*, Crabbé, J., Ross, E. J., and Thorn, G. W., *J. Clin. Endocrinol. and Metab.*, 18, 1159 (1958).

[6] Jenkins, D., Forsham, P. H., Laidlaw, J. C., Reddy, W. J., and Thorn, G., W., *Amer. J. Med.*, 18, 3 (1955).

[7] Genest, J., Nowaczynski, W., Loiw, E., and Sandor, T., Proc. Intern. Congr. of Biochem., Vienna, 1958 (in the press).

Effect of Age on the Heparin Content of Rat Skin

IN 1955, Shetlar and Masters[1] demonstrated a change with age in the composition of acid mucopolysaccharides of human cartilage. They found that both uronic acid and hexosamine decreased with age but the decrease in hexosamine was less striking, indicating a relative increase in some hexosamine-containing polysaccharide. Further evidence for a shift in the relative proportions of acid mucopolysaccharides in ageing cartilage was provided by Kuhn and Leppelmann[2], who reported a decrease in the ratio of galactosamine to glucosamine. Recently, Kaplan and Meyer[3] observed that the content of chondroitinsulphuric acid in human cartilage decreased and that of keratosulphate increased with age. On the basis of an increasing glucosamine to galactosamine ratio, Hallén[4] concluded that the ratio of keratosulphate to chondroitinsulphuric acid in human nucleus pulposus increased with age. A difference in the distribution of mucopolysaccharides between embryonic and mature pig skin was reported by Loewi and Meyer[5]. They found that hyaluronic acid comprised 78 per cent of the total skin mucopolysaccharides in the embryo and only 30 per cent in the adult, whereas chondroitinsulphuric acid-*B* represented 5–12 per cent in embryonic skin and 64 per cent of total mucopolysaccharides in adult skin.

The development of quantitative methods for extracting and separating mucopolysaccharides from tissues has made it possible to investigate the relationship of age and the qualitative and quantitative mucopolysaccharide content of skin. These studies lead to the isolation of heparin in relatively large amount from rat skin. Analysis of skin from rats of various ages demonstrated increased concentrations of all mucopolysaccharides in the very young animal. Most dramatic, however, were the changes in heparin concentration.

Acetone-dried skin from male Sprague–Dawley rats, ranging in age from 21 days to 2·6 years, was extracted by a modification of a method published previously[6]. The mucopolysaccharide mixture was precipitated by cetyl pyridinium chloride in the presence of 'Celite' as described by Korn[7]. Hyaluronic acid, the chondroitinsulphuric acids, and heparin, were extracted by successive washes with 0·3 *M*, 1·2 *M* and 2·1 *M* sodium chloride, respectively.

The results, presented in Table 1, are expressed as μgm. of uronic acid (as determined by the carbazole reaction[8]) per gm. of acetone-dry skin. The concentration of hyaluronic acid and chondroitinsulphuric acid in young rats was found to be approxim-

Table 1. EFFECT OF AGE ON CONCENTRATION OF MUCOPOLYSACCHARIDES IN RAT SKIN

Age (days)	Hyaluronic acid	Chondroitinsulphuric acid	Heparin
	(μgm. uronic acid/gm. dry rat skin)		
21*	979	385	421
23*	894	406	562
44*	1,232	337	388
57*	439	218	100
74*	540	197	92
217*	578	273	111
406	320	140	70
473	450	200	110
561	480	257	68
893	380	135	35
951	538	256	100

* These results were obtained from pools of 8–10 rat skins for each age group. In the older age groups, the results represent values from individual animals.

ately twice that in mature animals. The amount of heparin in young animals was surprisingly large. After 50 days of age the heparin concentration had diminished by 75 per cent. Thereafter, the level of all acid mucopolysaccharides in skin appeared to remain fairly constant.

In order to demonstrate unequivocally that the mucopolysaccharide present in the 'heparin' fraction was indeed heparin, 56 gm. of acetone-dried rat skin were extracted and the mucopolysaccharides separated as above. Approximately 9 mgm. of mucopolysaccharide were isolated from the 'heparin' fraction. The material was subjected to chromatography on 'Dowex 1 × 2', chloride resin, using increasing concentration of sodium chloride for elution. Heparin was removed with 2 *N* sodium chloride. Analyses showed hexosamine, uronic acid (by the carbazole reaction[8]) and ester sulphate to be present in a molar ratio of 1·0 : 1·6 : 2·0, respectively. The amino-sugar, determined by the method of Stoffyn and Jeanloz[9], was glucosamine. The substance had a specific rotation of + 90° and an anticoagulant activity identical to that of a commercial preparation of beef heparin (General Biochemicals, Inc.) with 136 units per mgm.

In view of the large number of mast cells in rat skin, the presence of heparin in this tissue is not surprising. Monkhouse *et al.*[10] reported heparin activity in extracts of rat skin but no chemical analyses were published, and their preparations contained a low order of anticoagulant activity.

Whether the heparin present in skin is entirely intracellular cannot be ascertained from the present data. It is conceivable that some heparin is secreted into the extracellular space during the life-cycle of the mast cell. This conjecture is consistent with a recent observation by R. S. Geiger and A. Dorfman (unpublished results) that granules are extruded from mast cells during culture of a mouse mastocytoma.

The finding of large amounts of heparin in skin and its relative decrease with age has many physiological implications. The role of heparin in lipæmia-clearing and blood coagulation suggests that changes in tissue concentration may influence these phenomena. It is possible that a local decrease in heparin affects deposition of fibrin or lipids.

A more detailed report of the methods and results will be published elsewhere.

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[1] Shetlar, M. R., and Masters, Y. F., *Proc. Soc. Exp. Biol. Med.*, 90, 31 (1955).

[2] Kuhn, R., and Leppelmann, H. J., *Liebigs Ann. Chem.*, 611, 254 (1958).

[3] Kaplan, D., and Meyer, K., *Nature*, 183. 1267 (1959).

[4] Hallén, A., *Acta Chem. Scand.*, 12, 1869 (1958).

[5] Loewi, G., and Meyer, K., *Biochim. Biophys. Acta*, 27, 453 (1958).

[6] Schiller, S., Mathews, M. B., Jefferson, H., Ludowieg, J., and Dorfman, A., *J. Biol. Chem.*, 211, 717 (1954).

[7] Korn, E. D., *J. Biol. Chem.*, 234, 1325 (1959).

[8] Dische, Z., *J. Biol. Chem.*, 167, 189 (1947).

[9] Stoffyn, P. J., and Jeanloz, R. W., *Arch. Biochem. Biophys.*, 52, 373 (1954).

[10] Monkhouse, F. C., MacKneson, R. G., and Bambers, G., *Proc. Soc. Exp. Biol. Med.*, 95, 489 (1957)

A Possible Function of Planktonic Photophores

A NUMBER of unrelated groups of planktonic animals have developed the curious arrangement of upwardly directed eyes combined with downwardly directed photophores; this has been described and discussed by Hardy[1], who has suggested a number of possible explanations.

Could this singular arrangement sometimes constitute a mechanism whereby the members of a vertically migrating species can keep together even when the most important factor, light from the surface, is brusquely interrupted?

Consider a gregarious species, mainly light-controlled, which occupies depths between 600 m. and 800 m. on a fine day, and between 200 m. and 400 m. by night. At midday after a sunny morning the animals at 600 m. will be comparatively well illuminated, but those at 800 m. may be almost at the threshold of perception of daylight. Now suppose that, as must not infrequently happen, the Sun is suddenly obscured by thick cloud for the rest of the day. The highest animals will start to swim up towards the levels of light-intensity that suit them best; the deeper animals, however, cannot at once do so, for they are now suddenly unable to perceive any daylight at all. Their upward-pointing eyes, however, may be able to perceive the diminution, in size and intensity, of the pattern of the photophores of their relatives above them, now rapidly climbing. Since they are gregarious, this situation might urge them in their turn to climb, so as to keep station with their fellows.

In this way, I suggest, it may be possible for a species to undertake light-controlled vertical migrations, even though the light may be acting on many of the individuals only at second hand.

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[1] Hardy, A. C., "The Open Sea", Part 1, "The World of Plankton", Chap. 13 (Collins, 1956).

PLANT PHYSIOLOGY

Interaction of Growth Inhibitor and a Natural Germination Stimulator in the Dormancy of *Fraxinus excelsior* L.

DORMANCY of seeds of the genus *Fraxinus* has been variously ascribed to immaturity of the embryo[1], mechanical resistance of the seed coats[2,3], and the presence of growth-inhibiting substances within the pericarp[3] and in the mucilage surrounding the embryo[4]. A recent study of dormancy in the fruits of *F. excelsior* L. has shown that these explanations are inadequate.

When the fruit of *F. excelsior* is shed from the tree, the embryo is small, but morphologically complete. Before germination occurs, the embryo grows by cell division and cell extension from little more than half, to the full length of the seed. This occurs most rapidly in moist storage at warm temperatures (18–20° C.) and is greatly retarded by the presence of the pericarp, which apparently restricts the oxygen supply to the embryo. Even after the embryo is fully grown, neither the fruits nor the naked seeds germinate unless after-ripened in moist storage at 5° C. for at least 4 months. Excised, unchilled embryos remain dormant indefinitely if precautions are taken to preclude leaching, whereas embryos excised from chilled seeds begin to germinate immediately.

An extensive survey of the changes in growth-promoting and growth-inhibiting substances has been carried out during embryo growth, chilling and germination. Diethyl ether extracts, separated into acid and neutral fractions, and aqueous extracts were concentrated and chromatographed on paper using as a developing solvent 80 parts of *iso*propanol to 20 parts of 0·15 *M* ammonia. The chromatograms were assayed for active substances using the *Avena* coleoptile straight-growth technique, and embryos excised from seeds of *F. excelsior*.

Ether extracts of ungerminated seeds contain no growth-inhibiting substances, but a growth-promoting substance, tentatively identified as indoleacetonitrile by means of its R_F value in various solvents and colour reactions, is present in ether extracts of fresh seeds and gradually disappears during growth of the embryo. Aqueous extracts of dry seeds contain no growth inhibitors, but on imbibing the seeds with water an active growth-inhibiting zone R_F value 0·6–0·8 appears in the chromatograms of extracts of endosperms and embryos imbibed 24 hr., and increases in activity during the following 48 hr. This inhibitor not only prevents the growth of *Avena* coleoptiles, cress roots and the germination of lettuce seeds, but also inhibits radicle growth of excised embryos of *F. excelsior*.

Although the seeds germinate after chilling, no reduction in activity of the inhibitor in either the endosperms or the embryos could be detected during or after the period of chilling. After the initial stages of germination, however, a very slight reduction in activity was detected, accompanied by the appearance of an ether-soluble inhibitor with similar properties to the β-inhibitor of Kefford[5].

Excised, unchilled embryos germinate rapidly if leached in distilled water for 48 hr., whereas embryos excised from leached intact seeds remain dormant. A very slight reduction in the level of inhibitor was detected in leached embryos, but not in the embryos excised from leached intact seeds. If leached (non-dormant) embryos are placed on filter paper moistened with inhibitor solution, very little growth ensues This was used as the basis for biological assays in which ten leached excised embryos were placed in the eluates from sections of chromatograms in place of the ten coleoptile sections normally used. It was shown that the fraction which prevented embryo growth was identical with the inhibitor of coleoptile growth (Fig. 1), and was active at all stages of after-ripening and germination of the seed.

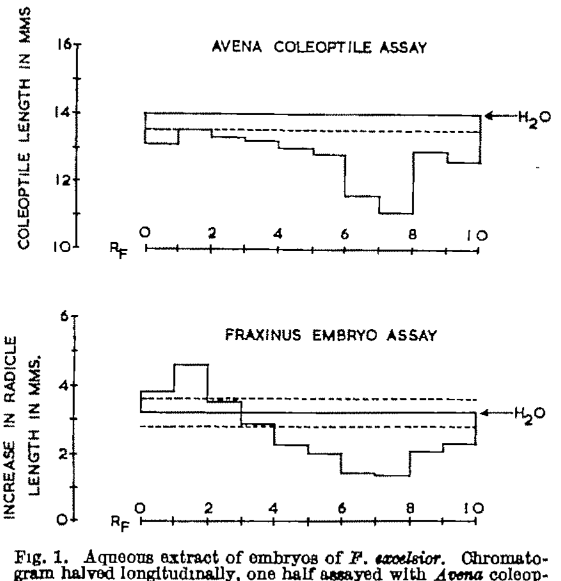

Fig. 1. Aqueous extract of embryos of *F. excelsior*. Chromatogram halved longitudinally, one half assayed with *Avena* coleoptile test, other half assayed with leached *F. excelsior* embryos

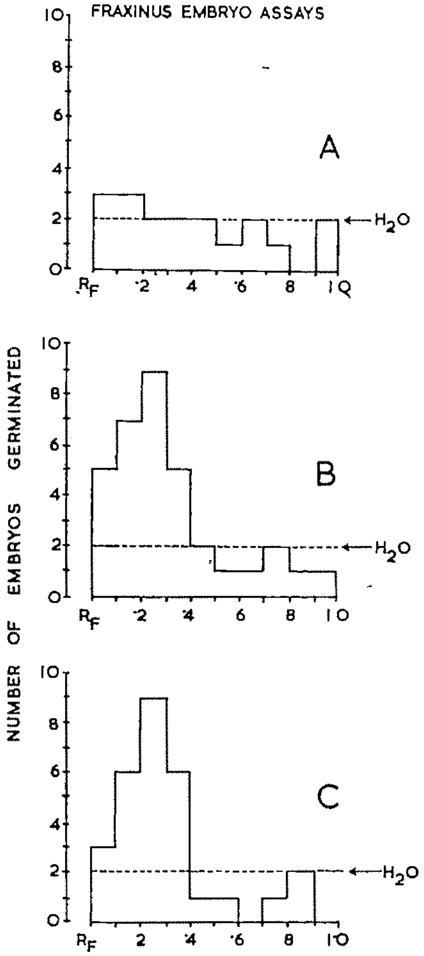

Fig. 2. Effect of aqueous embryo extracts on germination of unchilled embryos. *A*, Extract of unchilled embryos; *B*, extract of chilled, non-germinating embryos; *C*, extract of chilled, germinating embryos

Thiourea has the property of counteracting the effect of the inhibitor on the germination of lettuce seeds. Excised unleached (dormant) embryos of *F. excelsior* germinate readily if placed on filter paper moistened with 0·5 per cent thiourea solution, but no reduction in the level of inhibitor can be detected within the embryos.

As chilled embryos are able to germinate without leaching or thiourea treatment, and the process of chilling does not decrease the inhibitor content of endosperm or embryos, it was decided to investigate the possibility of the production of a germination-promoting substance. As no such substance could be demonstrated using the coleoptile straight-growth

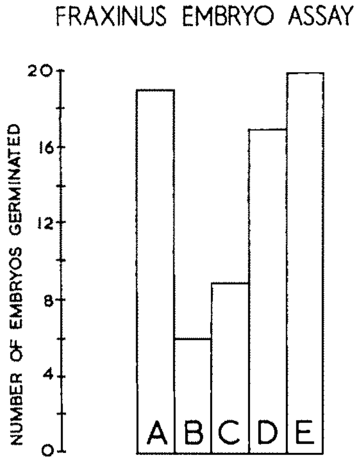

Fig. 3. Interaction between inhibitor and germination promoter. Leached, unchilled embryos on filter paper moistened as follows *A*, water only; *B*, inhibitor only; *C*, inhibitor + eluate from zone R_F 0·15–0 35 of extract of unchilled embryos; *D*, inhibitor + 0 5 per cent thiourea; *E*, inhibitor + eluate from zone R_F 0 15–0·35 of chilled embryos

Note.—Embryos from *A* produced stunted seedlings, those from *D* would not grow further, those from *E* produced normal seedlings

technique, unleached (dormant) embryos of *F. excelsior* excised from unchilled seeds were used in place of the coleoptile sections in biological assays. It was found that the zone R_F 0·15–0·35 in chromatograms of aqueous extracts of chilled, and of chilled germinating embryos, stimulated the germination of the dormant assay embryos within three days. This zone of stimulation was shown not to be present in extracts of unchilled embryos (Fig. 2), nor in the endosperms of seeds at any stage of after-ripening or germination.

That this germination-stimulating substance has the property of counteracting the effect of the inhibitor was demonstrated by the following experiment. The inhibitor-containing zone of a chromatogram (R_F 0·6–0·8) was cut into four strips and the strips moistened separately in Petri dishes with: (*a*) distilled water as a control; (*b*) eluate from zone R_F 0·15–0 35 from a chromatogram of extract of 10 unchilled embryos; (*c*) 0·5 per cent thiourea solution; (*d*) eluate from zone R_F 0·15–0·35 from a chromatogram of extract of 10 chilled embryos. A further control of filter paper moistened with distilled water (that is, no inhibitor) was prepared. Twenty unchilled, leached (non-dormant) embryos were placed in each of the above five dishes. It was found that there was little growth in the dishes containing either inhibitor alone, or inhibitor plus eluate from unchilled embryos, whereas in the dishes containing inhibitor plus eluate from chilled seeds, inhibitor plus thiourea, and distilled water alone, full germination took place within four days (Fig. 3).

The subsequent growth of variously treated embryos was investigated. Seedlings grown from excised chilled embryos, and excised unchilled embryos treated with the germination stimulator extracted from chilled embryos, were apparently normal when compared with those produced from normally germinated after-ripened seeds. Seedlings produced from unchilled embryos, germinated after leaching in water, were stunted, and embryos treated with thiourea did not produce seedlings, even after washing to remove excess thiourea.

Tests in which embryos in various stages were treated with synthetic gibberellic acid have shown that although the dormant embryos can be stimulated to germinate slowly by treatment with this substance, nevertheless the stunting of seedlings grown from unchilled embryos is not removed by gibberellic acid. It would seem, therefore, that the germination promoter is not gibberellic acid, a conclusion which is supported by the fact that small amounts of tissue may be used for extraction, in contrast with the large amounts used for the demonstration of natural gibberellins.

The fact that the inhibitors and germination stimulator may be detected by the embryos in which they occur naturally is good evidence that the results obtained *in vitro* reflect actual changes within the after-ripening seed. The evidence obtained suggests that the deep dormancy of seeds of *F. excelsior* is imposed by the presence of growth inhibitors in both endosperm and embryo During chilling, this dormancy is overcome by the production of a germination-stimulating substance in the tissues of the embryo.

A detailed account of the experimental work will be published elsewhere.

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[1] Lakon, G., *Naturwiss. Z. Forst- und Landw.*, 9, 285 (1911).
[2] Steinbauer, G. P., *Plant Physiol.*, 12, 813 (1937).
[3] Asakawa, S , *Bull. Gov. For Exp. Sta.*, *Tokyo*, 83, 29 (1956).
[4] Ferenczy, L., *Acta Biol.*, 1, 17 (1955).
[5] Kefford, N. P , *J. Exp. Bot* , 6, 129 (1955).

Effects of Gibberellic Acid on Excised Pea Roots

IN the course of an investigation of inter-varietal differences in the growth of excised roots of field peas (*Pisum sativum* L. var. *arvense*), some effects of gibberellic acid have been observed which seem worthy of brief record. Seeds were surface sterilized with 0·1 per cent mercuric chloride and germinated for two days. Root tips (5 mm.) were excised and grown in the dark at 25° C. in Petri dishes containing the inorganic medium of Bonner and Addicott[1] with 4 per cent sucrose, vitamin B_1 (0·1 mgm./l.) and nicotinic acid (0·5 mgm./l.). Ten tips were employed in each treatment, two being grown in each dish. After seven days the roots were measured and the apical 10 mm. of each was transferred to fresh medium for a further week.

Roots of the variety Minerva Maple grew extremely well (Table 1) and, so far as is known, no other variety of either field pea or garden pea has been found to grow as rapidly under similar conditions. By comparison, the growth of the variety Winter Maple was poor and it decreased markedly in the second passage. Gibberellic acid did not influence the growth of Minerva and in this respect this variety behaved in a similar way to the roots of intact cress seedlings[2]. With Winter Maple on the other hand, 0·5 and 25 mgm./l. gibberellic acid caused significant though small increases in growth in the first passage. The lower concentration stimulated growth again in the second passage but the higher concentration gave no stimulation. When seedlings of Winter Maple, which

Table 1. EFFECTS OF GIBBERELLIC ACID ON EXCISED ROOTS OF MINERVA MAPLE AND WINTER MAPLE

Variety	Concentration G A (mgm./l)	Length of root (mm)	
		Passage 1	Passage 2
Minerva Maple	0	77·6 ± 7·0*	87·5 ± 10·0*
	0·5	71·8 ± 4·0	90·6 ± 7·7
	25·0	89·0 ± 8·7	83·5 ± 9·5
Winter Maple	0	41·9 ± 1·8	15·7 ± 2·1
	0·5	48·8 ± 2·5	25·8 ± 1·1
	25·0	46·7 ± 1·7	14·7 ± 2·1

G.A , Gibberellic acid. * Standard error

is not a dwarf variety, were grown in an inorganic medium the roots grew much more rapidly than the excised roots in sterile culture. The above results may indicate that one of the factors limiting the growth of the excised roots of Winter Maple is a gibberellic acid-like substance.

The most striking effect of gibberellic acid on the excised roots, however, was on the development of root hairs. In most plants, root hairs are formed much more readily in moist air than in water[3]. In excised pea roots some hairs form on the tissue of the inoculum but the major part of the root is normally hairless. However, in the presence of gibberellic acid root hairs continued to develop in an acropetal manner, the youngest occurring on the tissue which had just completed extension growth. The covering of hairs was considerably denser at 25 mgm./l. than at 0·5 mgm./l. In Minerva the hairs were quite normal in appearance. In Winter Maple the hairs were shorter than in Minerva and at the higher concentration of gibberellic acid they tended to swell at the tip, the region of the hair which is generally believed to exhibit the greatest plasticity[3].

That gibberellic acid stimulates the normal development of hairs on excised roots raises the possibility that substances of this type may participate in the formation of these structures in intact plants.

The seeds employed in this work were kindly supplied by the Plant Breeding Institute, Cambridge.

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[1] Bonner, J., and Addicott, F. T., *Bot. Gaz.*, 99, 144 (1937).

[2] Brian, P. W., Hemming, H. G., and Radley, M., *Physiol. Plant.*, 8, 899 (1955).

[3] Cormack, R. G. H., *Bot. Rev.*, 15, 583 (1949).

Reversal of Genetic Dwarfism in *Tephrosia vogelii* by Gibberellin

IN conjunction with a breeding programme designed to produce plants of *Tephrosia vogelii* Hook f. with a higher rotenone content, multiple crosses were made between two lines of similar morphology, but of separate geographical origin. Nine dwarf plants were obtained as segregants in an F_2 population of 103 individuals. These dwarfs were characterized by greatly reduced internodes, hypocotyls, petioles, and rachises. Leaflets were also much smaller, and leaf colour was a darker green than normal. The number of leaves per plant, the number of leaflets on corresponding leaves, and the size and colour of the cotyledons were unaffected.

The potassium salt of gibberellic acid in an aqueous solution of 100 p.p.m. was applied in the form of a light spray to three of the dwarf plants. Three additional dwarf plants were sprayed with indoleacetic acid at 100 p.p.m., and the remaining three were sprayed with water as a control. The initial applications were made approximately one month after germination, and an additional spraying was given one week later. After responses to gibberellin were noted, two of the plants in this treatment were withheld from further applications, while one plant continued receiving weekly treatments of gibberellin.

Elongation of the petioles, rachises and leaflets was observed one week after the initial application of gibberellin. Leaves and internodes which had elongated prior to treatment were unaffected. Two weeks after the initial treatment with gibberellin, the three plants showed elongated internodes and leaves which were identical in appearance to those of normal seedlings. Normal growth continued in the plant which received additional periodical treatments of gibberellin. In those plants which were withdrawn from the gibberellin treatment after two applications, the dwarf condition reappeared gradually, and one month after the last treatment the plant was indistinguishable from the dwarf controls except for the elongated stem produced during the period of gibberellin treatment. These results indicate that the dwarf segregants of *T. vogelii* are completely dependent on periodic applications of gibberellin for normal growth.

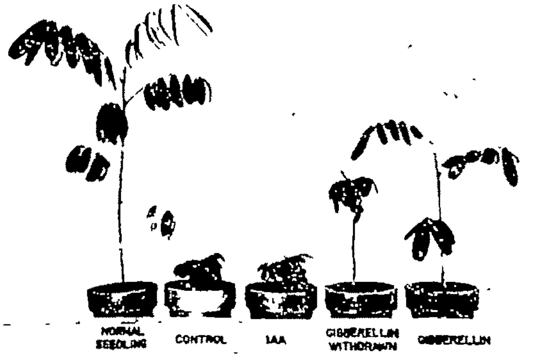

Fig. 1. Seedlings of *Tephrosia vogelii* Hook f showing a normal F_2 seedling, an untreated dwarf segregant, a dwarf treated with indoleacetic acid, a dwarf treated and then withdrawn from gibberellin, and a dwarf on continued gibberellin treatment

There was no growth response in plants treated with indoleacetic acid when compared to the controls. Fig. 1 shows (from left to right) a normal seedling of *T. vogelii*, an untreated dwarf segregant, a dwarf treated with indoleacetic acid, a dwarf treated with gibberellin for two weeks and then withdrawn, and a dwarf which has continued to receive periodical applications of gibberellin. A similar reversal of genetic dwarfism in other legumes[1] and in maize[2] has been reported.

We are indebted to Merck Sharp and Dome International, Rahway, New Jersey, for supplying the gibberellin used in this study.

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[1] Brian, P. W., and Hemming, H. G., *Physiol. Plantarum.*, 8, 669 (1955).

[2] Phynney, B. O., *Proc. U.S. Nat. Acad. Sci.*, 42, 185 (1956).

BIOLOGY

Water Hyacinth : a Curse or a Crop?

An organism often multiplies explosively when carried to a new environment. Rabbits in Australia are a familiar example, and now we have water hyacinth (*Eichornia crassipes*) in south-east Asia, the Nile, and the Congo[1-3]. In time, enemies of the invader will probably evolve in the new environment, or be introduced into it, and restore a balance, but that may take many years. In the meantime, there is disruption of old patterns of life and effort is therefore put into attempts to eradicate the invader and restore the *status quo*. This is obviously wise, but it is by no means certain that eradication will be successful, so that it may be prudent to try to find how best to live in the new circumstances. The invader may often be useful.

The main disadvantages of working with a weedy crop, or with wild vegetation, are that there is competition and products are harvested with widely different properties and potentialities. They would be useful separate but not mixed. Water hyacinth produces a pure stand. Acres of water that hitherto carried little plant-life now carry a rich growth. This makes the plant resemble a crop rather than a weed and raises a fundamental issue. Hitherto, agricultural research has been based on the idea that we know what crops we want and then grow them in increasing abundance in increasingly exacting environments. Biochemical engineering introduces the opposite outlook : it strives to find what an area will produce most abundantly and then looks for a use for it[4].

A floating plant is ideally adapted for mechanized harvesting but possible uses for water hyacinth have not been intensively sought. Cattle will eat it, as Davies[3] pointed out, and Hora[5] describes its use as pig food in south-east Asia. It has been fermented to give methane[6] and compost[7]. When harvested it contains 95 per cent of water. This is a disadvantage for many uses, but is a great advantage if it should prove possible to use the leaf as a source of extracted protein, because general experience suggests (cf. ref. 8) that protein extraction is easier from wet than from dry leaves. Leaf protein extraction is now being studied in several places with many different types of leaf, but the basic principle of extraction is always the same and is very simple[9] ; the fresh leaf is pulped and the juice, neutralized if necessary, is separated. It carries with it much or most of the protein, and this is coagulated and filtered off as a dark green cake which generally has little flavour after washing with water. It can be used as it stands in various dishes that are reasonably palatable[10], or a stable protein can be made by extracting the chlorophyll and lipids with a solvent. So many species have now given satisfactory yields of protein that there is good reason to expect that it could be extracted from water hyacinth. But this has not, apparently, yet been demonstrated.

This communication is a plea for some re-orientation of thinking in areas suffering from, or threatened with, water hyacinth. There is a case for directing some research towards finding uses for the pest instead of concentrating all efforts on its eradication. The practicability of those mentioned here has not been established and difficulties may arise with some of them. But the quantity of water hyacinth is now so large, and so much effort is already being put into dragging it from waterways, that it would seem reasonable to set up a unit to study all the possibilities.

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[1] Gay, P. A., *Nature*, 182, 538 (1958).

[2] Gay, P. A., and Berry, L., *Geographical J.*, 125, 89 (1959).

[3] Davies, H. R. J., *Nature*, 184, 1085 (1959).

[4] Pirie, N. W., *Research*, 10, 20 (1957)

[5] Hora, S. L., *Science and Culture*, 17, 231 (1951).

[6] *Research*, 5, 522 (1952)

[7] Basak, M. N., "Water-Hyacinth Compost", Pub. Government of West Bengal (1948).

[8] Pirie, N. W., "Ann. Rev. Plant Physiol.", 10, 33 (1959).

[9] Pirie, N. W., *Food Manufacture*, 32, 416 (1957) ; *Nutrition*, 12, 3 (1958).

[10] Pirie, N. W., *Lancet*, ii, 961 (1959).

Chick Feeding in the Wandering Albatross *Diomedea exulans* Linnæus

It has long been known that the young of the wandering albatross (*Diomedea exulans*) do not fly until eight or nine months after hatching, and the problem of how this long fledgeling period fits into the breeding cycle of the adults has caused some speculation among naturalists acquainted with the species[1-6]. Excellent descriptions are available of the birds at sea and on the breeding grounds, but it is only recently that detailed study within a large population has been undertaken[7].

As early as 1929 it was suggested that individuals of the species may not breed each consecutive season because of the necessity of feeding the young throughout their first winter[1]. Such a phenomenon was held by Murphy[2] to be highly improbable. It was maintained that the only hypothesis consistent with the few other available winter observations at breeding stations[8] was that the chicks, after being fed for the first three or four months, were deserted by their parents, who departed to sea to prepare for the following breeding season. The nestlings, it was stated, went for the last four months, throughout their first winter, without feeding but surviving off stored body reserves of visceral and subcutaneous fat until fully fledged and able to leave the nesting grounds, shortly after the adults had returned again to breed.

Studies of the related royal albatross (*Diomedea epomophora*) have since demonstrated a bi-annual breeding cycle in which the young are fed throughout the fledgeling period of 236 days[4]. Since Matthews's original description[1], winter feeding in the wandering albatross has been noticed at three other stations. In the Antipodes Islands eight-month-old chicks were receiving meals[9] and at Kerguelen feeding has been seen during August and September[5]. On Gough Island, adults were observed with eight-month-old chicks some time before the return of breeding birds[10].

Recent workers at South Georgia[7] spent five months in the field during October 1958–March 1959, most of it on Bird Island 54° 00′ S. ; 38° 02′ W., the densest breeding ground of the wandering albatross in the world.

The albatross returns to South Georgia in mid-November ; but before the return of the main body of breeding birds single adults were observed paying brief visits to nestlings of not more than 10 min. duration. Banding operations began at Bird Island on November 26 and during the following month 656 nestlings were banded. In the course of this work, nestlings being handled frequently regurgitated large fresh meals and the sight of adults feeding young was fairly common. There is no direct evidence that these adults were in fact feeding their own young; but nestlings were often seen begging from adults which refused them. Conversely, there is no indication in the royal albatross that adults feed random chicks[4].

In adult wandering albatrosses there is a significant difference in body-weight between the sexes, but at the time of departure the few young examined were not necessarily lighter or heavier than the adults. The range of body-weights in twelve juveniles a few days before leaving varied between 5·6 and 11·3 kgm. while that of 105 adults of both sexes was 6·35 to 9·15 kgm.

Table 1. WEIGHTS OF ADULT WANDERING ALBATROSSES, BIRD I

Sex	No	Mean (kgm)	Standard deviation (kgm.)	Standard error (kgm)	Range (kgm.)
Male	52	9·11	0 60	0 083	8 20–9 15
Female	53	7 27	0 52	0·071	6 35–7 30

The lightest of the juveniles had been losing weight at the rate of 95 gm./day during the preceding twelve days while the mean daily loss of the heaviest over eighteen days was 200 gm./day (Fig. 1). The combined mean daily loss in four juveniles during the three weeks preceding departure was 162·5 gm./day (No. 64, *S.D.* 3·30, *S.E.* 0·412, range 1–650 gm.).

Thus a young albatross leaving the breeding grounds in December with a flight weight of 9 kgm. must, if it had fasted the whole winter, have weighed over 23 kgm. the previous July—more than twice the body-weight of an adult male. Rankin[3], calculating from daily weight increments of four chicks in the month following hatching, estimated that the weights at the end of June would be between 30 lb. 12 oz. and 25 lb. 13 oz. (13 9–11 8 kgm.). In comparison with adult weights these appear to be reasonable estimates, but they are strictly comparable with those of fully fledged juveniles preparing to depart in December. Thus they are no evidence for the hypothesis that they are put forward to support, rather the reverse, for young albatrosses of such peak weights would not have survived a winter without fairly regular feeding.

The young of most of the procellariiformes reach peak weights in excess of the parent body-weights.

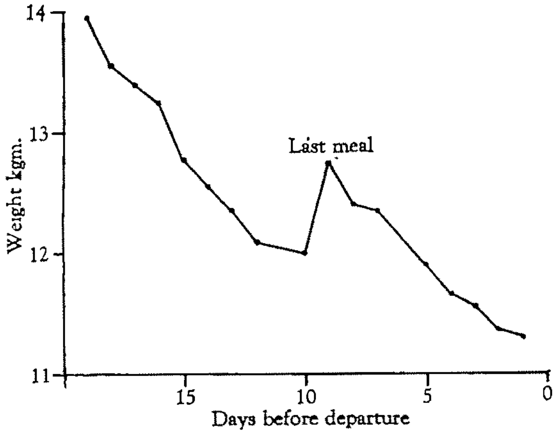

Fig. 1. Weight of a young wandering albatross during its last weeks at the nest

In some species there may be desertion by adults but recent studies have demonstrated that it is more usual for the young to lose surplus weight during the latter part of the fledgeling period while they are still being fed[4,11–15]; although this feeding is less frequent than during the earlier part of the chick period.

While nothing can yet be deduced of the feeding regime of the wandering albatross, the evidence now available clearly indicates that the young are fed throughout the fledgeling period. The 'starvation' hypothesis is therefore untenable and it must be held that the wandering albatross, like the royal albatross, breeds only once every two years.

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[1] Matthews, L. H , "Birds of South Georgia", *Discovery* Reports, 1, 567 (1929).

[2] Murphy, R. C , "Oceanic Birds of South America", 561 (Amer. Mus. Nat Hist., New York, 1936).

[3] Rankin, N., "Antarctic Isle", 167 (Collins, London, 1951).

[4] Richdale, L. E., "The Post-Egg Stage in Albatrosses" (Dunedin, New Zealand, 1952).

[5] Paulian, P., "Pinnipedes, Cetacés, Oiseaux des Iles Kerguelen et Amsterdam", *Mem. de l'Inst. Scient. de Madagascar*, A, 8 (1953).

[6] Jameson, W , "Wandering Albatross" (Hart-Davis, London, 1958).

[7] Tickell, W. L. N , and Cordall, P. A., "South Georgia Biological Expedition 1958/'59", unpublished report to F I D S. (1959).

[8] Hutton, F. W , *Ibis*, 1, 276 (1865)

[9] Turbott, in Richdale 1952 (ref. 4, *loc. cit.*)

[10] Holdgate, M., "Mountains in the Sea" (Macmillan, London, 1958)

[11] Richdale, L. E , *Trans. Roy. Soc , N.Z.*, 73, Parts 2 and 3, 97, 217 (1943)

[12] Richdale, L. E., *Trans Roy Soc. N Z.*, 73, Pt. 4, 335 (1944), 74, Parts 1 and 2, 32, 165 (1944).

[13] Richdale, L E , *Emu*, 43, 191 (1944).

[14] Richdale, L. E., *Emu*, 43, 24 (1943)

[15] Tickell, W. L. N., "The Dove Prion", F I.D Scientific Report No 33 (in the press)

Hydrophilic Gels for Biological Use

PLASTICS to-day enjoy wide use in many fields, and it is natural that the possibility of their employment in permanent contact with living tissues has been seriously considered. A study of the literature shows that almost all known plastics have been proposed for this purpose at various times. The question of structural similarity with the tissue has not, however, been emphasized although physiologically unfavourable effects were observed in most cases of application of normal type plastics.

The extensive application of plastics for alloplastic and prosthetic uses requires solution of the problem of compatibility of the material with the live tissue. It is indisputable that non-biological material represents a foreign body having contact surfaces showing large deviations from normal physiological behaviour.

The result of poor compatibility is either an immediate severe local irritation or a tumorous growth which sometimes appears only after a longer interval The cause of these phenomena, as has been ascertained, is to be found among other factors in the mechanical irritation or in the long-term slow diffusion of low molecular irritant substances from the plastic itself, so that every plastic must be judged from this point of view. All high polymers used so far in medicine are impermeable to metabolites. The majority of them are hard solids so that they can cause mechanical irritation. Further, one must expect a certain slow leaching of irritating substances from them. Polyvinyl alcohol is an exception; but it has the major disadvantage that it is absorbed.

The demands to be placed on a suitable plastic after consideration of the above-mentioned effects are thus: (1) a structure permitting the desired water content; (2) inertness to normal biological processes (including resistance to the degradation of the polymer and to reactions unfavourable to the organism); (3) permeability for metabolites.

Materials with these properties must have hydrophilic groups. Further, they must have a three-dimensional structure with at least enough cross-linkages to prevent absorption.

Of a large number of plastics we found co-polymers of glycolmonomethacrylate with several tenths per cent of glycoldimethacrylate to be the most suitable. They hydrolyse only with difficulty, even under severe conditions, and are indifferent to biological materials. A further advantage is that their mechanical properties and water content can be adjusted to meet requirements. They withstand heat sterilization without damage.

Hydrophilic materials of a marked polyelectrolyte character (with basic groups, or with carboxyl groups) have unfavourable effects on the organism, swell excessively and their chemical stability does not attain the stability of glycol esters.

Gels based on glycolmethacrylates are also noteworthy for their transparency (in the case of polyglycolmonomethacrylate with a water content less than 30 per cent and with the higher glycols at all concentrations). The water content, and thus the shape of the object, is, after attaining equilibrium at the given temperature, constant and depends on the structure and number of cross-linkages. The appropriate monomers can be prepared by the alcoholic re-esterification of methylmethacrylate with glycol followed by selective extraction of the di- and mono-esters from the reaction mixture. The desired shapes are prepared by polymerization of aqueous solution in suitable moulds. Rubber-like and soft gels can be stiffened by polyamide fibres either in the form of flock or threads. Before use it is necessary thoroughly to leach out the shapes.

The formation of a porous structure by the rapid polymerization of glycolmonomethacrylate is of great interest. With a water content greater than corresponds to the equilibrium capacity of the gel, a porous structure will arise by coalescing of the water-phase droplets into interconnected channels. The size of the pores depends on the polymerization conditions. On drying the porous sample, a transparent solid material is formed which takes on the original shape and structure on immersion in water.

After embedding in the living organism they do not show any irritating reactions as do other materials. Histological sections show successful encapsulation of the inserted object. Detailed biological results will be published in due course.

After more than four years of testing, these materials have now passed through the stage of application such as filling after enucleation of the eye. Promising results have also been obtained in experiments in other cases, for example, in manufacturing contact lenses, arteries, etc.

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Origin of Placental Trophoblast

IN 1949 I published a hypothesis suggesting that the cyto- and syncytio-trophoblast were derived from the granulosa cells that surround the egg when it leaves the ovary; the argument, however, depended on indirect evidence, and although this indirect evidence has further accumulated it is not proposed to discuss it here[1].

Since then the work initiated by Barr and Bertram[2] on the nuclear sex chromatin has provided an opportunity to test the hypothesis. Obviously, if the trophoblast is fœtal its sex will always correspond with that of the fœtus; if it is maternal it should always be female. Workers have studied the normal placenta, chorionepithelioma and the hydatidiform mole. With respect to the normal placenta it must be recognized that my hypothesis refers only to the syncytium and the cytotrophoblast, and when this latter tissue disappears, only to the former. Other tissues in the placenta, such as those forming the vascular and supporting structures of the villus and cells derived from the outer covering of the blastocyst (chorionic epithelium), are undeniably of fœtal origin, and perhaps the last-named tissue also gives rise to cells that may equally in current usage be called cytotrophoblast. The only valid evidence, therefore, will result from sexing of the syncytium.

Park[3] worked on the embryos in the Carnegie Institution. His slides were old and not specially stained. He claimed that the trophoblast was always of the same sex as the fœtus; but when certain syncytial cells were female when they should have been male, he argued that these particular ones must have been derived from maternal stroma blending with developing trophoblast.

Klinger[4], Parmentier[5], Stevenson and McClarin[6], Bohle and Hienz[7] either did not examine the syncytium or found it unsuitable for study.

With respect to chorionepitheliomata Park[8], Oosterhuis and Levy[9] seem to have ignored the syncytium and the former suggests that trophoblast of chorionepitheliomata may not show any reliable sex difference.

Klinger and Ludwig[10] have recently described a more satisfactory method of staining the sex chromatin body and have applied it to the hydatidiform mole[11]. The results are interesting, for of nine moles examined all were female. The authors, in explanation, suggest (1) only female embryos develop moles; (2) there may have been a nuclear change, that is, the nuclei of the mole were polyploid, or (3) moles are derived from the maternal tissues. The first two explanations cannot be correct, for one of their moles developed with a male embryo, and Klinger and Schwarzacher[12] state that polyploid male nuclei (even $8n$) only occasionally show a sex chromatin mass and even so it is distinguishable from the average female diploid. The third explanation, and in my view the correct one, they do not elaborate.

In view of the results of experiments on homotransplantation it is strange that the syncytiotrophoblast is accepted so readily as of fœtal origin. Medawar[13] initiated the work showing that homotransplants do not take if the donor and recipient are not of identical genetic constitution, and the human mother and fœtus are generally not so identical. Tolerance can be produced in mice to homologous skin grafts only by manipulations in the recipient around the time of birth[14]. The human mother has not only more highly developed immunological processes but also can in no sense be considered new-born, or a twin of the fœtus. Medawar suggests that the mammalian fœtus is usually protected from the consequences of homotransplantation as it has its strictly independent circulation. However, the human syncytiotrophoblast has a relationship with the maternal circulation that could not be closer. Algire *et al.*[15] can grow homografts indefinitely by inserting such into containers that prevent vascularization by the host and entry of cells. May the syncytium be Nature's analogue to Algire's millipore filter ?

It appears thus that the normal syncytium and the chorionepithelioma are difficult to sex but that the hydatidiform mole is female. It is considered that this supports my earlier suggestion that the syncytium is derived from granulosa cells, and its function is to protect the fœtus from the immunological processes of the mother.

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[1] Gordon, I., *Lancet*, i, 807 (1949).
[2] Barr, M. L., and Bertram, E. G., *Nature*, 163, 676 (1949).
[3] Park. W. W., *J. Anat.*, 91, 369 (1957).
[4] Klinger, H. P., *Acta Anat.*, 30, 371 (1957).
[5] Parmentier, R., *Bull. fed. soc. gyn. et d'obst.*, 9, Supp., 30 (1957).
[6] Stevenson, A. C., and McClarin, R. H., *Nature*, 180, 198 (1957).
[7] Bohle, A., and Hienz, H. A., *Klin. Woch.*, 35/36, 981 (1956).
[8] Park, W. W., *J. Path. and Bact.*, 74, 197 (1957).
[9] Oosterhuis, W. W., and Levy, I. S., *Ned. Tydschr. v. Geneesk*, 101, 901 (1957).
[10] Klinger, H. P., and Ludwig, K. S., *Stain Tech.*, 32, 235 (1957).
[11] Klinger, H. P., Ludwig, K. S., Schwarzacher, H. G., and Hauser, G. A. *Gynaecologia*, 146, 328 (1958).
[12] Klinger, H. P., and Schwarzacher, H. G., *Nature*, 181, 1150 (1958).
[13] Medawar. P. B., *Quart. J. Micr. Sci.*, 89, 239 (1948).
[14] Billingham, R. E., and Brent, L., *Phil. Trans. Roy. Soc.*, 242, 439 (1959)
[15] Algire, C. H., Weaver, J. M., and Prehn, R. T., *Ann. N.Y. Acad. Sci.*, 64, 1009 (1957).

Influence of Filming and of Surface Texture on the Settlement of Marine Organisms

WHEN a well-cleaned surface is allowed to stand in sea water it becomes coated with a film in a few hours. The film increases in thickness for at least the first few days after it has formed. Several reports are to be found in the literature of a filmed surface being more favourable to settlement than a freshly cleaned one[1-4]. It has been widely assumed that this is true for all marine larvæ, since a permanent surface suitable for settlement is bound to collect a film in the sea. The extreme view has indeed been expressed that filming may be a necessary pre-requisite to the attachment of marine larvæ, although larvæ of some sedentary organisms have been observed to attach themselves to newly submerged surfaces before any visible film has formed[2].

A second assumption frequently made is that marine larvæ will attach themselves to a roughened surface more readily than to a smooth one.

Both these assumptions are being currently tested on a variety of invertebrate larvæ. Small rectangular pieces of chemically inert material are used, the upper surfaces of which are prepared in a series of grades of roughness from a shiny glossy finish to a coarsely ground surface. The surfaces are filmed for periods of 24 hr. or more and arranged in a factorial experiment around the periphery of a cylindrical dish containing larvæ, and slowly rotated in order to equalize such external influences as uneven illumination, to which the larvæ might be susceptible.

Table 1. NUMBER OF LARVÆ OF *Spirorbis borealis* AND OF *Bugula flabellata* SETTLING ON EQUAL AREAS OF PREPARED CHEMICALLY INERT SURFACES

Type of surface texture	*Spirorbis borealis*			*Bugula flabellata*		
	Filmed	Clean	Total	Filmed	Clean	Total
Glossy	826	23	849	175	967	1,142
Smooth but dull	273	239	512	93	1,327	1,420
Rough	1	0	1	88	960	1,048
Very rough	11	0	11	54	596	650
Totals	1,111	262	1,373	410	3,850	4,260

Our experiments, which will be fully reported elsewhere, have already shown that neither of the above generalizations is valid. The larvæ of many organisms settle preferentially on smooth rather than on rough surfaces, while others are indifferent to surface texture. Furthermore, although the majority of larvæ appear to choose a filmed surface, those of one species at least, *Bugula flabellata*, settle more readily on a clean unfilmed surface than on a filmed one. Moreover, we have verified that the film eschewed by *Bugula flabellata* was not a film of an accidentally unfavourable character, since it simultaneously attracted the larvæ of another animal, *Spirorbis borealis*, known for some time to prefer filmed surfaces[3]. The two kinds of larvæ in roughly similar numbers were placed together in a dish containing a series of surfaces, both filmed and unfilmed. As can be seen from the results in Table 1, larvæ of *Spirorbis* were attracted but those of *Bugula* were repelled by the film. Furthermore, *Spirorbis* larvæ clearly avoided the rougher surfaces, while *Bugula* larvæ were almost indifferent to surface texture.

It seems clear that a considerable diversity of behaviour towards films exists even in these simple ciliated larvæ ; they seem able to respond both to the form of the surface and also to the presence or absence of a film overlying it. Further work may well show that many of them can distinguish qualitatively between films, as the larvæ of *Ophelia bicornis* appear to do[4]. No simple generalizations therefore seem applicable to the reactions of settling larvæ on filmed surfaces.

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[1] ZoBell, C. E., and Allen, E. C., *J. Bact.*, 29, 239 (1935). Scheer, B. T., *Biol. Bull., Woods Hole*, 89, 103 (1945). Cole, H. A., and Knight-Jones, E. W., *Fish. Invest., Lond.*, 2, 17, No. 3 (1949). Daniel, A., *J. Madras Univ.*, 25. 189 (1955).
[2] Miller, M. A., Rapean, J. C., and Whedon, W. F., *Biol. Bull., Woods Hole*, 94, 143 (1948).
[3] Knight-Jones, E. W., *J. Mar. Biol. Assoc., U.K.*, 30, 201 (1951).
[4] Wilson, D. P., *J. Mar. Biol. Assoc., U.K.*, 34, 531 (1955).

Elminius modestus Darwin on the Isle of Cumbrae (Firth of Clyde)

THE distribution and rate of spread of the immigrant intertidal barnacle *Elminius modestus* Darwin in the British Isles and in the rest of north-west Europe have been summarized recently[1]. The most northerly published record is that of Connell[2], who found a single specimen of *Elminius* on Farland Point, Isle of Cumbrae, on March 7, 1955 ; the nearest published records to this are those for Stranraer and Kirkcolm in Loch Ryan[1], about 55 miles to the south.

In view of the interest attached to the well-documented[1] rate of spread of this barnacle, search has been made at intervals since 1955 for further specimens on the Isle of Cumbrae; but none was seen until September 3, 1959, when two specimens were found on the exposed tip of Keppel Point, about 525 m. north-east of Connell's find. The two *Elminius*, which were left *in situ*, had settled about 3 m. apart, near mean tide-level, on rock among large *Balanus balanoides*; both specimens were small (3·5 and 3·6 mm. mean diameter, September 29) and presumably had settled during the summer of 1959.

Since it is believed that cross-fertilization is obligatory in *Elminius* and that in consequence individuals separated by a distance exceeding 5 cm. cannot breed[1], much larger settlements are required before the species can become fully established here.

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[1] Crisp, D. J., *J. Mar. Biol. Assoc. U.K.*, 37, 483 (1958).
[2] Connell, J. H., *Nature*, 175, 954 (1955).

Mass Rearing of Barnacle Cyprids in the Laboratory

Interest in laboratory experiments on ships' fouling organisms is increasing, yet attempts to rear barnacle larvæ to cyprid stage have generally met with only limited success[1–6]. It therefore seems appropriate to place on record a reliable rearing method, which has been developed here to provide material in connexion with experiments on the use of ultrasonic antifouling equipment. The barnacle mostly used was *Elminius modestus*, as this important fouling organism produces nauplii all the year round. As a result of many series of experiments the following simple technique was evolved.

All sea water used is first pasteurized by heating to 60° C. and then cooled to room temperature. Masses of ripe embryos are dissected out of adult barnacles and placed in a dish of sea water on the side remote from the light. Stage I nauplii soon hatch from a proportion of the egg masses and swim rapidly to the lighted side of the dish, tending to leave behind Protozoa that may have been introduced with the egg masses. From the lighted side the nauplii are pipetted into 1 or 3 l. 'Pyrex' Berzelius beakers of sea water to give a larval density not exceeding 300 per l. *Skeletonema costatum* (Plymouth Collection No. 106), cultured in standard Erdschreiber medium[7], is used as food. Actively growing culture is added at a density of 200–500 cells per mm.^{3} at the beginning of the experiment. The diatoms tend to settle to the bottom, but gentle stirring with a glass rod once a day is generally found to resuspend them sufficiently, particularly as the nauplii are able to feed on them while they are on the bottom. The vessels are covered and kept on a laboratory bench where they receive north daylight. No change of water or vessel is carried out and growth of the food plant usually makes good the quantity consumed, but excess food should always be present and more diatoms are added whenever necessary. Considerable fluctuations of temperature are tolerated. At a temperature of about 22° C. cyprids of *Elminius* appear after six days and have been obtained at all months of the year.

More than 90 per cent of the stage I nauplii eventually reach cyprid stage in such a culture and 80 per cent or more of these settle if provided with a suitable substratum. These figures contrast with the overall survival to the settled state of 16·3 and 12·7 per cent recently reported for individually reared larvæ of *Balanus eburneus* and *B. amphitrite* var. *denticulata*, respectively[4,5]. The main reasons for the failure of previous workers to obtain consistently high yields are probably contamination of cultures with other organisms, especially ciliates, overcrowding or stale or wrong food.

Although gentle bubbling of air from a glass jet has often been used to help keep the food suspended, careful experiments show that results with *Elminius* in bubbled vessels are no better than in stagnant cultures where satisfactory conditions have been established. There is thus no support for the idea recently put forward[8] that agitation of the water is necessary for ecdysis to be successful. Similarly, penicillin and streptomycin, which have been recommended in culturing oyster larvæ[9], do not produce significantly better results when added to healthy cultures, although they help in poor culture conditions where many larvæ have died.

Besides *Elminius* the cyprids of *Verruca stroemia*, *Chthamalus stellatus*, *Balanus amphitrite* var. *denticulata*, *B. perforatus*, *B. balanoides*, *Acasta spongites* and *Pyrgoma anglicum* have all been reared in large numbers. The larvæ of the two latter species are being described. Other plant cultures have been used as food with varying success and their relative values are being tested. Already it is clear that *Elminius* grows and survives better when fed on *Skeletonema* than on *Phaeodactylum*. Japanese workers have also found *Skeletonema* suitable as food for barnacle larvæ[10]. *Chthamalus stellatus*, however, though capable of being reared satisfactorily on the small flagellate, *Isochrysis galbana*, has not been reared successfully on *Skeletonema* and therefore seems to show a difference from Balanidae with which the fine setulation of the larval limbs in *Chthamalus* is probably correlated. The use of liver or egg extract as food[11] has not been tried, but would obviously need a procedure entailing frequent water changes to remove decomposing food.

I am grateful to the Marconi International Marine Communication Company, Limited, for financing this research, to Dr. Mary Parke of the Marine Laboratory, Plymouth, for supplying the plant cultures and to Prof. E. W. Knight-Jones for his advice and criticism.

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[1] Groom, T. T., *Phil. Trans. Roy. Soc.*, 185, 121 (1894a).
[2] Bassindale, R., *Proc. Zool. Soc. Lond.*, 1, 57 (1936).
[3] Sandison, Evvor E., *Trans. Roy. Soc. S. Africa*, 34, 69 (1954).
[4] Costlow, jun., J. D., and Bookhout, C. G., *Biol. Bull.*, 112, 313 (1957).
[5] Costlow, jun., J. D., and Bookhout, C. G., *Biol. Bull.*, 114, 284 (1958).
[6] Barnes, H., and Barnes, Margaret, *Canad. J. Zool.*, 37, 15 (1959).
[7] Gross, F., *J. Mar. Biol. Assoc. U.K.*, 21, No. 2, 753 (1937).
[8] Barnes, H., and Barnes, Margaret, *Limnol. and Oceanogr.*, 3, No. 1, 29 (1958).
[9] Walne, P. R., *J. Mar. Biol. Assoc. U.K.*, 37, 415 (1958).
[10] Hudinaga, M., and Kasahara, H., *Zool. Mag. (Japan)*, 54, 108 (1941).
[11] Howie, D. I. D., *Nature*, 181, 1486 (1958).

Terrestrial Ostracods of New Zealand

TERRESTRIAL ostracods have been known since 1953 from South Africa when Dr. R. F. Lawrence found specimens of *Mesocypris terrestris* Harding, in the humus of the Knysna forest[1]. Although there is no published record they have been found on several occasions in New Zealand, also in forest litter. Specimens from Leith Saddle, Dunedin, have recently been extracted alive by slowly drying out litter in a Berlese funnel, with a water-jacket maintained at a temperature of 40° C. and with the end leading into water, not alcohol. The temperature must be low to prevent the ostracods closing their valves and remaining immobile, and this is probably the reason why they have been found so infrequently. The animals are easily kept alive in a small dish lined with moist filter paper, and with a few fragments of humus to provide food and shelter.

The New Zealand specimens also appear to belong to the genus *Mesocypris* Daday, but differ in several respects from other described species. The valves of the shell are of equal size. Typical dimensions are: length, 0·56 mm., and height, 0 28 mm., the colouring being pale lemon unevenly distributed. Two specimens of the fifty which I have seen are considerably larger, being as much as 1·04 mm. long and 0·62 mm. high. The colour is also different, with patches of light brown. No males have yet been found. There is a marked dorso-ventral flattening, and the shell is moderately hairy. The appendages are broadly similar to those of *M. terrestris* with well-developed claws and a reduced number of setæ. A full description will be published later.

Locomotion is by means of the second antennæ and the third legs, both of which are armed with claws at the tips. An extra leg, used also for balance in steep places, is provided by the two halves of the furca, which act together. The animals are capable of moving up and down vertical slopes of humus, and one specimen was seen to climb several centimetres up the smooth sides of a glass dish. The first antennæ are used for sensory perception and balance, and with the setæ spread out explore objects ahead of the animal. They maintain a constant up and down motion, each alternating with the other. Sudden intense illumination causes the valves to close while bright light leads to rapid wandering in search of shelter. It is surprising to find apparently normal eyes, as Harding states that *M. terrestris*, like most litter-dwelling animals, appeared to be blind.

The Dunedin specimens were collected in ordinary forest litter from podocarp-broadleaf forest on Leith Saddle at an altitude of 1,200 ft. in places of no more than ordinary dampness. Experiments in the laboratory have shown that they are able to survive quite marked drying-out. although their activity decreases considerably and they tend to remain in holes in the humus. On the addition of more water they emerge and are capable of moving with great speed. On the other hand total immersion does not harm them and they will survive for at least a week. It is interesting that terrestrial Harpacticoid copepods were also found in the same habitat.

Other specimens known to me have been collected from Mount Arthur at 3,200 ft. (J. T. Salmon; 1946), Stewart Island (R. K. Dell and B. A. Holloway; 1955), Mount Egmont in sub-alpine forest (M. P. Büchler; 1954), at the foot of Mount Burnett, Nelson (R. Chapman; 1956), beech litter from Silverstream (J. D. Stout), Little Barrier Island 800 ft. (A. Chapman; 1959).

Further studies are in progress on the ecology and distribution of this New Zealand terrestrial ostracod.

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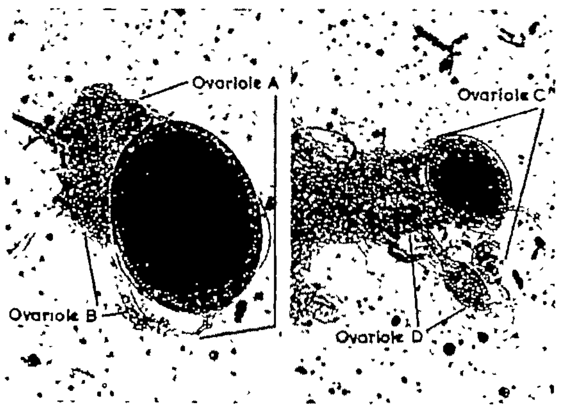
[1] Harding, J. P., *Ann. Natal Mus.*, 12, 359 (1953).

Ovaries of *Glossina morsitans*

CORRELATED with the viviparous type of reproduction found in the tsetse flies, the essential features of which were worked out as early as 1895 by Bruce[1], there is a great reduction in the number of ovarioles. Stuhlman[2] and Roubaud[3] studied the anatomy of the female reproductive organs and considered that each ovary consisted of a single ovariole, a view which has been perpetuated by every subsequent author. This communication, however, reports that in *Glossina morsitans* dissections have proved that each ovary contains, in fact, two ovarioles.

Dissections of teneral females were carried out in 0·9 per cent saline on a microscope slide. The ovarian sheaths were stripped off with a fine needle and the two ovarioles in each ovary released (Fig. 1, *a*, *b*). The slide was then drained of the excess saline and the ovaries mounted directly in a drop of polyvinyl-lactophenol containing a little lignin pink. This mountant produced a permanent preparation and avoided distortion of the ovarioles.



(*a*) (*b*)

Fig. 1. (*a*) Right ovary of a 24-hr.-old *Glossina morsitans*, dissected to show the two ovarioles. (*b*) Left ovary, ditto

In a study of the ovulation cycle of female *G. palpalis*, Helen Mellanby[4] showed that the first egg is ovulated from the right ovary, and the subsequent eggs alternately from left and right ovaries throughout the reproductive life of the fly. The cycles in *G. morsitans* and *G. swynnertoni* have been shown to be the same[5]. In the light of the fact that there is a total of four ovarioles, however, the ovulation cycle in *Glossina* requires a more detailed study. Fig. 2 shows the four ovarioles completely removed from the ovaries of a 24-hr. old female *G. morsitans*. The most advanced egg follicle is in one (*A*) of the two ovarioles in the right ovary; the ovum and yolk occupy about the lower third of the follicle. The

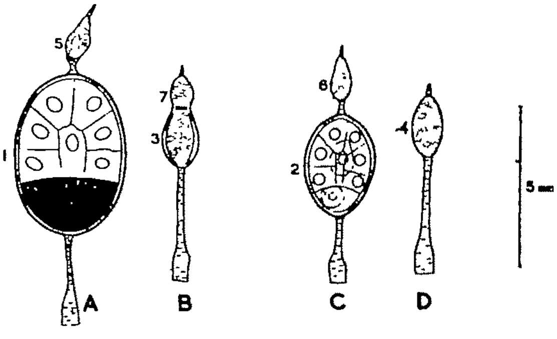


Fig. 2. The four ovarioles of a 24-hr.-old *G. morsitans*. Nos. 1–7: follicles in the order of development

next most advanced follicle is in one of the ovarioles (*C*) in the left ovary. This follicle shows only the beginning of yolk deposition. The second ovariole in the right ovary (*B*) shows the early stages in the development of its first egg follicle and the second ovariole in the left ovary (*D*) shows only the germarium and no signs of follicle development. From this appearance of the ovarioles in the newly emerged female it would seem that the general outline of the ovulation cycle as described by Helen Mellanby is true but must now be modified. The first egg is probably ovulated from ovariole *A*, the second from *C* and the third and fourth from *B* and *D*, respectively. The fifth egg produced will develop in the second follicle in ovariole *A*.

Further studies of the ovulation cycle in *G. morsitans* are in progress and will include those with particular reference to the state of the follicular tubes after ovulation.

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[1] Bruce, D., "Preliminary Report on the Tsetse Fly Disease or Nagana, in Zululand" (Bennett and Davies, Durban, 1895). (Original not seen; quoted from Buxton, P. A., "The Natural History of Tsetse Flies" (London School of Hygiene and Tropical Medicine, Memoir 10 (1955).)

[2] Stuhlman, F. *Arb. Kaiserl. Gesundheits.*, 26, 301 (1907). (Original not seen; quoted from Buxton, P. A., "The Natural History of Tsetse Flies", London School of Hygiene and Tropical Medicine, Memoir 10 (1955).)

[3] Roubaud, E., thesis No. 1344, University of Paris (1909).

[4] Mellanby, H., *Parasitol.*, 29, 131 (1937).

[5] Mellanby, K., *Parasitol.*, 29, 142 (1937).

VIROLOGY

Native Protein, an Immunizing Agent against Virus Infection

WITH several plant virus diseases, an infection, after becoming generalized, often results in apparent recovery of the plants, followed by an acquired immunity from reinfection[1]. The term, acquired immunity, has further been applied to the interference between virus strains in cross-protection tests.

The prevailing theory is that the virus particles are attached to 'acceptor points' in the plant cell[2]. Once these 'acceptors' are occupied no new virus particles of the same virus or related strains (requiring same 'acceptor') can multiply in the cell.

In this preliminary communication, a kind of immunization, apparently not due to antagonism of related strains, is reported.

Purified tobacco mosaic virus was prepared from tobacco plants, inoculated by a single lesion of common tobacco mosaic virus. The virus was purified by differential centrifugation[3]. This virus preparation, containing about 1 per cent tobacco mosaic virus, was degraded by Fraenkel-Conrat's method, using 67 per cent acetic acid[4]. Any undegraded virus was removed by ultra-centrifugation. The native protein obtained was tested on *Nicotiana glutinosa* L., and found to be free of virus particles.

In the first series of experiments half sides of detached leaves of *N. glutinosa* were inoculated with this native protein solution, freshly prepared, using 'Carborundum' as an abrasive. The other half of the leaf was similarly rubbed with water. The leaves were kept on moist filter paper, in Petri dishes. After varying intervals the whole leaves were rubbed with a diluted solution of tobacco mosaic virus; 'Carborundum' was again used. After 3–4 days lesions appeared and were counted separately for each half-side. Results, a mean of eight replicates, are summarized in Table 1.

Table 1. EFFECT OF NATIVE PROTEIN ON TOBACCO MOSAIC VIRUS INFECTION IN DETACHED LEAVES (AVERAGE OF EIGHT REPLICATES)

No. of inoculations with native protein	Tobacco mosaic virus inoculation, days after last protein treatment	No. of lesions in protein treated/untreated half-leaves	Ratios of lesion numbers in protein treated/untreated half-leaves (average)
1	1 hr.	52 : 46	1·15
1	1 day	52 : 56	0·96
1	2 ,,	60 : 81	0·79*
1	4 ,,	36 : 59	0·61*
2 (second after 24 hr.)	2 ,,	69 : 120	0·59*
,,	4 ,,	16 : 57	0·36†
2 (second after 2 days)	2 ,,	19 : 47	0·39†
,,	4 ,,	31 : 56	0·57*

* Significant at 0·05 per cent. †, Significant at 0·01 per cent.

Due to the extent of variation in lesion numbers within the replicates, the ratio between the two leaf half-sides was found to be much more constant, and was therefore used for statistical analysis.

As shown, no effect was found if the leaves were inoculated with tobacco mosaic virus either immediately or 24 hr. after the virus protein had been applied. The effect of the protein is more pronounced from two doses than from one, and increased somewhat after 4 days as compared with after 2 days.

In a second series similar results were obtained. The number of lesions and the ratio between treated and untreated half-sides varied; in some cases a ratio of even 1 : 9 in the protected leaf half-side was obtained.

In further experiments undetached leaves of *N. glutinosa* plants were treated. Each formula was tested on two plants—eight half-sides altogether. Protein and tobacco mosaic virus inoculations were conducted as described above. Results, a mean of eight replicates, are summarized in Table 2.

It should be added that the absolute number of lesions, as well as the ratio values, should be regarded cautiously, as the susceptibility of leaves and plants may vary. Nevertheless the striking effect of the proteins is clearly evident.

These preliminary results suggest the formation of 'protective substances' within the plant. As no

Table 2. EFFECT OF NATIVE PROTEIN ON TOBACCO MOSAIC VIRUS INFECTION IN UNDETACHED LEAVES (AVERAGE OF EIGHT REPLICATES)

No. of inoculations with native protein	Tobacco mosaic inoculation days after last protein treatment	No. of lesions in protein treated/untreated half-leaves	Ratios of lesion numbers in protein treated/untreated half-leaves (average)
1	1 hr.	38 . 37	1·12
1	1 day	46 : 39	1·20
1	2 days	30 . 44	0·69*
2 (second after 24 hr.)	2 ,,	39 : 49	0·64*
2 (second after 2 days)	4 ,,	38 : 76	0 56†

* Significant at 0 05 per cent. † Significant at 0 01 per cent.

result was obtained when the protein inoculations were done immediately before tobacco mosaic virus infection, direct inhibiting effect does not seem to be the cause. A direct inhibiting effect of native protein, if mixed with tobacco mosaic virus before inoculation, is known to occur, but only in concentrated doses[6]. With dilute solutions as used in our experiments no inhibition could be observed[5].

The increase in the depressing effect observed after two protein applications, and after 4 days, as compared to 1 and 2 days, seems to suggest an analogy with antibody formation in animals.

Attachment of the protein to 'acceptor points' appears unlikely, as the protein itself does not reproduce. After infection the virus multiplies and establishes itself thoroughly before cross-protection is effective[7]. The initial amount of protein applied would, therefore, not be sufficient to produce this effect.

A full report will be published elsewhere.

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[1] Price, W. C., *Phytopath.*, 26, 503 (1936).
[2] Bawden, F. C., "Plant Viruses and Virus Diseases", 122 (Chronica Botanica Co., 1956).
[3] Steere, R. L., "Adv. Virus Res.", 6, 57 (1959)
[4] Fraenkel-Conrat, H., *Virol.*, 4, 1 (1957).
[5] Bawden, F. C., and Pirie, N. W., *J. Gen. Microbiol.*, 17, 80 (1957).
[6] Fraenkel-Conrat, H., and Williams, R. C., *Proc. U.S. Nat. Acad. Sci.*, 41, 690 (1955).
[7] Salaman, R. N., *Phil. Trans. Roy. Soc.*, B, 229, 137 (1938).

Multiplication of Vaccinia Virus in Serum-free and Serum-containing Cell Cultures

THE use of tissue culture cells for the production of virus preparations and vaccines is increasing and will gain prominence in the future. A report has recently appeared concerning the possible use of cell cultures for the standardization of smallpox vaccines[1]. At present the cell lines used for the production of virus vaccines are grown in media which contain of necessity serum of one species or another[2]. Several cell lines which grow in media free of serum have been reported[3], and the advantages that these cells might have for vaccine production are obvious. Their use, however, could well be limited by their ability to support virus growth to a high titre. This report describes a comparison of vaccinia growth in serum-free and serum-containing mouse lung cells.

The cells used for growth studies were isolated from (new-born) white Swiss mice by the method of Younger[4]. The serum-containing line proliferated rapidly on a growth medium consisting of 10 per cent horse serum and 90 per cent medium 199[5]. The serum-free daughter culture was adapted to rapid growth on a medium in which the serum was replaced by 1 per cent Difco 'Bactopeptone' and 50–100 mgm. per cent dextrose[3]. The serum-containing line has been designated *MSI*, and the peptone line *MPDI*. At the time of use the *MSI* line was in its one hundred and twelfth passage (2¼ yr.) and the *MPDI* line in its sixty-fifth passage in peptone (1¾ yr.).

The vaccinia virus stock preparation strain *IDH–E*[6], which was normally maintained in chick embryos, was passed once in *MSI* or *MPDI* cells prior to experimental use. This was done to circumvent the possibility that any of the chick embryo protein might be carried along as a stimulatory factor when the virus was added to the cell cultures.

The *MSI* or *MPDI* cells were inoculated into a volume of 15 ml. of medium in 'Pyrex' milk dilution bottles, the total number of cells being approximately 3×10^5 (2×10^4 cells per ml.). After 48 hr. of incubation at 37° C. the cells were inoculated with 0·1 millilitre of a $1{\cdot}0 \times 10^{-3}$ dilution of the stock virus preparation. Immediately after inoculation, and every 24 hr. for a total of 8 days, 0·4 ml. of the supernatant fluid was harvested and stored frozen. Following the final harvest the virus was titrated by inoculating 0·1 ml. of a ten-fold dilution into each of 4, 16 mm. × 150 mm. test-tubes containing $2{\cdot}5 \times 10^5$ cells and 1 ml. of medium. The cell types used in this titration were, *MPDI*, *MSI* or N.C.T.C. clone 929 (strain *L*)[7]. The tubes were incubated at 37° C. at an angle of 5° and examined daily by use of a microscope. The titration end-point was considered as the final tube showing at least 20 per cent cell destruction within 10 days. The 50 per cent tissue culture infective dose ($TCID_{50}$) was then calculated by the method of Reed and Muench[8]. Plaques regularly appeared in 3–6 days in tubes which eventually showed the necessary cell destruction.

As an alternative method of titration, rabbits were inoculated intradermally with 0·1 ml. of the same ten-fold dilutions of virus. The end-point in this titration was taken as the highest dilution of virus eliciting an inflammatory lesion which progressed through pustulation and scar formation.

The results of a typical growth study are shown in Table 1. Only the titre of the 5-day harvest is shown, since the rate of growth was approximately the same in both cell lines. The maximum titre was usually reached on the fifth or sixth day, and the peak titre was maintained in both cell lines for at least 24 hr. The greatest amount of virus present in the peak titres in both cell lines averaged about 10 times higher than that previously shown with the Levaditi strain of vaccinia in serum-containing mouse lung cells[9].

It is apparent that cells grown in the absence of serum or its derivatives are able to support vaccinia multiplication to about the same degree as serum-containing lines. It also appears that serum-free cells are not as sensitive for the determination of

Table 1 MULTIPLICATION OF VACCINIA IN SERUM-FREE AND SERUM-CONTAINING CELL LINES. MAXIMUM TITRES

Virus grown in :	Negative $\log_{10}$ $TCID_{50}$ titre of virus titrated in : *MSI*	*MPDI*	*L*	Rabbits
MSI (serum)	6 3	6 0	6 5	6·3
MPDI (serum-free)	6 0	5 5	6 0	6·2

vaccinia infective titres as are serum-containing lines. This, however, may be related to the decreased stability of small numbers of virus particles in a non-serum-containing medium. The important point in this study is that the final titre of vaccinia is similar in both cell lines. After four passages in serum-free cells the virus has not yet shown a greatly diminished infectivity for rabbits as has been reported in other serum-containing lines[1].

Experiments are planned using other viruses in this same cell system. We are indebted to Dr. Wm. Cassel for provision of the vaccinia virus used in these studies.

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[1] Cutchins, E., and Warren, J., *Proc. Soc. Exp. Biol. Med.*, **97**, 456 (1958).

[2] Hilleman, M. R., *et al.*, *J. Amer. Med. Assoc.*, **163**, 4 (1957). Salk, J. E., *ibid.*, **158**, 1239 (1955).

[3] McQuilkin, W. T., Evans, V. J., and Earle, W. R., *J. Nat. Cancer Inst.*, **19**, 885 (1957). Waymouth, C., *ibid.*, **22**, 1003 (1959). Pumper, R. W., *Science*, **128**, 363 (1958).

[4] Younger, J. S., *Proc. Soc. Exp. Biol. Med.*, **85**, 202 (1954).

[5] Morgan, J. F., Morton, H. J., and Parker, R. C., *Proc. Soc. Exp. Biol. Med.*, **73**, 1 (1950).

[6] Cassel, W. A., *Virol.*, **3**, 514 (1957).

[7] Sanford, K. K., Earle, W. R., and Likely, G. D., *J. Nat. Cancer Inst.*, **9**, 229 (1948).

[8] Reed, L. J., and Muench, H., *Amer. J. Hyg.*, **27**, 493 (1938).

[9] Pumper, R. W., *Amer. J. Hyg.*, **69**, 79 (1959).

SCIENTIFIC METHOD

A Proposal to replace Belief by Method in the Pre-mensural Sciences

NOT only in his posthumous work, "Fundamental Theory"[1], but also in many of his earlier writings, Eddington was concerned to define the mathematical structure of what he called a 'measurable', and to distinguish a measurable from an observable, and an observable from the most primitive entity, which he regarded as resembling a geometrical point except that, instead of having position only (a far from primitive attribute), it has existence (or non-existence) only.

Eddington's proposed algebraic representations of these three concepts can be translated into the language of communication theory as follows: an entity has two characteristics, existence and non-existence, and so can be represented by one binomial unit of information (bit). An observable is represented by two entities and has four characteristics, while a measurable is represented by two observables (or four entities) and has sixteen characteristics. This means that an entity has one way of not existing out of two possibilities, an observable three out of four, and a measurable fifteen out of sixteen.

We have devised[2] an information model which represents the concept of a 'comparable', intermediate between an observable and a measurable. A comparable is represented by three bits of information, has eight characteristics, and accordingly it has seven ways of not existing within this universe of discourse.

The information structure (called the O-structure, because it incorporates what is necessary for measuring or 'observing'—Eddington's Z) is a hierarchical structure. If the whole structure consists of n bits there will be $\frac{1}{2}n$ in the ground-level, $\frac{1}{4}n$ in the next, and so on for the L levels, where $L = \log_2 n$, the number of bits in any level, l, being $n/2^l$.

So long as a science is based upon measurables, its primitives must be measurable things, that is, atoms, but if a science is based upon comparables, this allows comparisons to be made between an observable entity, such as a semantic relationship with an observable, such as a physiological response. Luria's work[3] along these lines makes meaning observable, but not measurable in the physical sense, and so, in principle, separates meaning from knowledge in just the same way as the cybernetic engineer separates information from meaning.

It is suggested that these separations now make it possible to formulate a scientific methodology of pre-mensural science, based upon similarity and comparable with the formal methodologies of dynamics and wave mechanics in the mensural sciences.

We represent the primitive entity, first by the binomial unit of information for ideal representation, and secondly as a two-way switch, in order that this ideal representation can be exemplified, at least in principle, by a cybernetic artefact. This double representation has been found capable of representing some characteristics of neural events, but owing to the great complexity both of these events and of experienced events (memories) it is not to be expected that detailed isomorphism can be established. However, it is easy to show how a connected system of switches can represent what is required, at least in a general way. If we start by assuming that all the connected switches have an equal probability of being open or closed, that is, a half-probability of existence, this will represent a system in which information or probability flow is everywhere maximal and uniform. If we now consider that some instability creates a linked succession of switches in the closed position, this will represent maximal existence and zero information. Such a linking inevitably produces a number of informationally isolated systems of zero existence and zero information; such systems represent events that are not being experienced. If we adopt Eddington's proposal that numbers between 0 and 1 represent degrees of statistical existence (phase) this allows us to suppose slight imperfections in our two types of system. The linking of chains of switches in the state of maximal existence may be imperfect (synaptic gaps), and as regards the isolated events that provide the possibility of recognition, these may be supposed to accumulate sufficient information by similarity to reach what might be called a 'semantic threshold' as a unit entity. It is such a unit entity that we envisage as the third element of a comparable, the other two being provided, in the case of perception, by two micro-neural events or quanta that together constitute an observable.

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[1] Eddington, A. S., "Fundamental Theory" (Camb. Univ. Press, 1946).

[2] *Com. Inst. Study Mental Images* 1, Part 1, 1 (1957). Gregory, C. C. L., and Kohsen, A., "The O-Structure: an Introduction to Psycho-physical Cosmology" (I.S.M.I., 1959).

[3] Luria, A. R., and Vinogradova, O. S., *Brit. J. Psychol.*, **50**, 2, 89 (1959).

FORTHCOMING EVENTS

Monday, January 11

Institution of Electrical Engineers, Electronics and Communication Section (at Savoy Place, London, W.C.2), at 5 30 p m.—Mr. G. G. Gourlet and Mr. G. F. Newell: "A Quadrature Network for Generating Vestigial-Sideband Signals"; Prof. D. G. Tucker: "The Input Impedance of Rectifier Modulators"; Mr. D. P. Howson and Prof. D. G. Tucker: "Rectifier Modulators with Frequency-Selective Terminations, with particular reference to the Effect of Even-Order Modulation Products".

Society of Chemical Industry, Surface Activity Group (joint meeting with the Oils and Fats Group, at 14 Belgrave Square, London, S.W.1), at 5.30 p.m.—Dr. C. G. Sumner: "Emulsions in Theory and Practice".

Manchester Literary and Philosophical Society (in the Reynolds Hall, Manchester College of Science and Technology, Sackville Street, Manchester), at 5.45 p.m.—Dr. G. E R. Deacon, F.R.S.: "Scientific Exploration of the Ocean" (Clayton Memorial Lecture).

Institute of Physics (at 47 Belgrave Square, London, S.W.1), at 6 p.m.—Prof. J. G. Wilson: "The Problem of Primary Cosmic Radiation".

Royal Geographical Society (at 1 Kensington Gore, London, S.W.7), at 8.30 p.m.—Mr. W. H. Melbourne and Mr. P. Garrard: "Explorations on the Bolivian–Peruvian Border".

Tuesday, January 12

Institution of the Rubber Industry (at the Royal Society of Tropical Medicine and Hygiene, Manson House, 26 Portland Place, London, W.1), at 5.30 p.m.—Mr. J. Westhead. "Chemical Plant Lining". 7 p.m.—Dr. J. N. Milne: "Hypalon Coatings".

Wednesday, January 13

Society for Applied Bacteriology (in the Barnes Lecture Theatre, Royal Society of Medicine, 1 Wimpole Street, London, W.1), at 9.45 a.m.—Winter Meeting. At 2.15 p.m. (joint meeting with the Microbiology Group of the Society of Chemical Industry)—Symposium on "Methods for the Evaluation of the Antibacterial Activity of Surface Active Compounds".

Bone and Tooth Society (in the Cuthbert Wallace Room, Royal College of Surgeons, Lincoln's Inn Fields, London, W.C.2), at 11 a.m.—Meeting on "Radioactive Materials and the Skeleton".

Physical Society, Low Temperature Group (in the Lecture Theatre of the Royal School of Mines, Prince Consort Road, London, S.W.7), at 4 30 p.m.—Dr. Audrey U. Smith: "Effects of Sub-Zero Temperatures on Living Cells, Tissues and Animals".

Institution of Electrical Engineers, Supply Section (at Savoy Place, London, W.C.2), at 5.30 p m.—Mr. E. C. Rogers and Mr. D J. Skipper. "Gaseous Discharge Phenomena in High-Voltage D.C. Cable Dielectrics".

Society of Chemical Industry, Corrosion Group (at 14 Belgrave Square, London, S.W.1), at 6 p.m.—Mr. J. N. Wanklyn. "Corrosion Problems of High Temperature Pressurized Water Reactors".

British Institution of Radio Engineers (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C 1), at 6.30 p.m.—Dr. G. T. Wright: "A Proposed Space-charge-limited Dielectric Triode".

Women's Engineering Society (at "Hope House", 45 Great Peter Street, London, S.W 1), at 7 p.m.—Mr. B. C. Godbold: "Health Physics and Nuclear Power".

Wednesday, January 13—Thursday, January 14

British Coal Utilization Research Association (at the Research Station, Randalls Road, Leatherhead, Surrey)—Symposium on "Aerodynamic Capture of Particles".

Thursday, January 14

Royal Society of Arts, Commonwealth Section (at John Adam Street, Adelphi, London, W.C.2), at 2.30 p.m.—Mr. R. L. E. Dreschfield: "The Preservation of Game in East Africa".

Institution of Electrical Engineers, Utilization Section (at Savoy Place, London, W.C.2), at 5 30 p m.—Mr. I. F. Davies and Mr. D. Dunthorne: "The Application of Power Transistors to the Operation of Gas Discharge Lamps from Direct Current Supplies".

Chemical Society (in the Lecture Theatre, The Royal Institution, Albemarle Street, London, W.1), at 7 30 p.m.—Prof C. Kemball: "Progress in the Study of Heterogeneous Catalysis" (Tilden Lecture).

Friday, January 15

Institute of Navigation (at the Royal Geographical Society, 1 Kensington Gore, London, S W.7), at 5 15 p.m.—Mr. J. F. Green and Mr. A. P. Glenny: "Heading Definition in Commercial Aircraft".

Society of Chemical Industry, Fine Chemicals Group (at 14 Belgrave Square, London, S.W 1) at 6.30 p.m.—Dr. A. H. Beckett: "Recent Developments in Analgesics".

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Mr. C. V. Dadd: "Biological Factors in Agricultural Aviation".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Assistant Biochemist, Basic Grade (with a science degree, preferably with honours in biochemistry or chemistry, or the A.R.I.C.)—The House Governor, The General Hospital, Birmingham 4 (January 16).

Physicist (Basic Grade), for duties in connexion with radiotherapy and isotopes—The Personnel Officer, St. Thomas's Hospital, London, S.E.1 (January 16).

Vice-Principal (with high academic and/or professional qualifications with substantial administrative experience of a relevant kind, for example, in education, industrial research or other professional work)—The Registrar, College of Advanced Technology, Gosta Green, Birmingham 4 (January 16).

Biologist, Assistant Experimental Officer grade (with a pass degree in a biological subject or equivalent), for work on plant virus diseases—The Secretary, Glasshouse Crops Research Institute, Worthing Road, Rustington, Littlehampton, Sussex (January 20).

Lecturer in Education—The Principal, Borough Road College, Isleworth, Middlesex (January 23).

Lecturer (Grade II) in Pure Mathematics—The Registrar, The University, Bristol 8 (January 25).

Technical Editor (Experimental Officer, preferably under 31, with at least H.N.C. or Inter-B.Sc. or equivalent, some knowledge of terms used in the fields of fuel and power, the ability to translate French and German, and preferably a knowledge of Russian) at the Ministry of Power, H.Q., London, for abstracting scientific articles; preparations of surveys of current literature; briefs and memoranda on fuel and power subjects—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting A.572/9A (January 29).

Lecturer in Mathematics at Victoria University of Wellington, New Zealand—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (New Zealand, January 31).

Lecturer (with some industrial or research experience, and preferably some teaching experience) in Physics—The Registrar, Royal Technical College, Salford 5, Lancs (January 31).

Senior Lecturer or Lecturer (preferably with a medical degree) in the Department of Pathology, University College of the West Indies—The Secretary, Senate Committee on Colleges Overseas in Special Relation, University of London, Senate House, London, W.C.1 (February 8).

Lecturer (with a special interest in plant physiology) in Botany at the University College of Rhodesia and Nyasaland—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (February 10).

Research Fellow (with M.Sc. or equivalent, with a special knowledge and/or experience of agronomy of tropical crops) in Tropical Crop Production at the Agricultural Research Station, University College of Ghana, Kade, in the forest region of Ghana—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (February 10).

Research Fellows (2) (with research experience in animal virology or in biochemistry) in the Department of Microbiology, John Curtin School of Medical Research, Australian National University, to work on either biological or biochemical aspects of animal virology—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (February 12).

Imperial Chemical Industries Research Fellows in Physics, Chemistry, Biochemistry, Engineering, Metallurgy and Pharmacology or any related subjects—The Registrar, The University, Liverpool (February 13).

University Librarian—The Registrar (Room 22, O R.B.), The University, Reading (February 13).

Professor and Chairman (with extensive experience in both teaching and research, preferably in the large animal field) of the Department of Animal Science—The Dean, Faculty of Agriculture, McGill University, Macdonald College, P Q., Canada (February 15).

Senior Lecturer in Geography at the University of Melbourne, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (February 15).

Senior Lecturer in Archæology and Ethnology at the University of Cape Town, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa, February 15).

University Tutor or Assistant Lecturer (medical and/or science graduate) in Microbiology—The Secretary, The Queen's University, Belfast (February 15).

Professor of Inorganic Chemistry—The Registrar, University Registry, Oxford (February 18).

Lecturer/Senior Lecturer (with good academic qualifications in electrical engineering, experience in the power field, and a working knowledge of electronic techniques) in Electrical Engineering at the University of Sydney, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia, February 19).

Pasture Utilization Officer (honours graduate in science or agricultural science (or equivalent qualifications), with some years postgraduate research experience and training in animal physiology and nutrition) in the Division of Plant Industry, Commonwealth Scientific and Industrial Research Organization, Canberra, A.C T., Australia, to participate in the current programme of investigation which seeks to understand the plant, animal and plant–animal factors in normal grazing systems, by studying the animal factors affecting the nutrition demands of the ewe (and lamb) at pasture, of the ability, or otherwise, of the pasture to satisfy these demands—Chief Scientific Liaison Officer, Australian Scientific Liaison Office, Africa House, Kingsway, London, W.C.2, quoting Appointment No. 130/891 (February 26).

Plant Breeder (honours graduate in science or agricultural science (or equivalent qualifications) with two or more years relevant postgraduate research experience) in the Division of Plant Industry, Commonwealth Scientific and Industrial Research Organization, Canberra, A.C.T., Australia, to collaborate with the established team of geneticists and plant breeders, and to assist in the extension of the current programme of studies on pasture plants by initiating independent studies on the breeding of pasture plants—Chief Scientific Liaison Officer, Australian Scientific Liaison Office, Africa House, Kingsway, London, W.C.2, quoting Appointment No. 130/403 (February 26).

Chair of Botany, tenable in the Newcastle Division of the University of Durham—The Registrar, University Office, 46 North Bailey, Durham (February 29).

Chair of Geography in the University of Canterbury, Christchurch, New Zealand—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (New Zealand, February 29).

Chemist (with a good honours degree, experience in crop and soil analysis, and an interest in field experimentation), to investigate anion-cation relationships in soils and crop nutrition—The Secretary, Rothamsted Experimental Station, Harpenden, Herts (February 29).

Imperial Chemical Industries Research Fellows in Chemistry, Biological Chemistry, Engineering Pharmacology, Physics, or related subjects—The Registrar, The University, Bristol (March 1).

Imperial Chemical Industries Research Fellow in Chemistry or Physics—The Secretary, The University, Exeter (March 7).

Imperial Chemical Industries Research Fellows in Chemistry, Physiological Chemistry, Agricultural Chemistry, Agricultural Botany, Horticulture, Microbiology, Geology, Physics, or related subjects—The Registrar (Room 22, O.R.B.), The University, Reading (March 7).

Assistant, Grade B (with a good degree in chemistry and preferably some teaching experience) to teach Chemistry to G.C.E. Advanced and Part 1 B.Sc. level—The Clerk to the Governors, South-West Essex Technical College, Forest Road, Walthamstow, London, E.17.

Assistant Master (bachelor) to teach Mathematics—The Headmaster, Wrekin College, Wellington, Salop

Assistant or Associate Professor (Ph.D. or equivalent) in the Soil Science Department, for duties which will include research, teaching and extension in the field of soil chemistry and soil fertility—Head of the Soil Science Department, University of Alberta, Edmonton, Alberta, Canada.

Biochemist (with a first- or second-class honours degree) in the Biochemistry Department, for duties mainly concerned with radioactive isotopes, including thyroid hormone synthesis and utilization—The Secretary, Glasgow Royal Infirmary, 54 Castle Street, Glasgow, C.4.

Chief Technician in the Department of Chemistry, for duties which will include the supervision of laboratories and technical staff, as well as responsibilities for furnishing and equipping new laboratories planned for completion in 1962—The Registrar, The University, Liverpool 3, quoting Ref. No. 240N

Editorial Assistant (science graduate, preferably in biochemistry or chemistry, and preferably with some scientific editorial experience) at the Editorial Office of the *Biochemical Journal*—The Secretary to the Editorial Board, *Biochemical Journal*, The Lister Institute of Preventive Medicine, Chelsea Bridge Road, London, S.W.1.

Electronic Engineer (with at least H.N.C. or equivalent and several years relevant experience), to design and build equipment for a research group—The Administrator, Clarendon Laboratory, The University, Oxford.

Entomologist (with a good honours degree in zoology or entomology, and preferably postgraduate research experience in forest or timber entomology) with the West African Timber Borer Research Unit, Kumasi, Ghana, to undertake studies of the biology, ecology and control of timber and lumber pests—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.197/510/01/C.1.

Head (with a degree in metallurgy, chemistry or a related science at the Ph.D. level or equivalent training with a number of years experience in the field of metal corrosion) of the Corrosion Section, Physical Metallurgy Division, Department of Mines and Technical Surveys, Canada, to initiate and supervise research and investigations on fretting corrosion, stress corrosion and corrosion fatigue of various metals and their alloys—Department 1517, National Employment Service of Canada, 61 Green Street, London, W.1.

Horticultural Advisory Officer (with a university degree in horticulture and postgraduate experience) in Malta, to develop cultivation of out-of-season flowers, fruit and vegetables primarily for export—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD 63/19/07/T.

Lecturer (graduate physical chemist or metallurgist) in Extraction Metallurgy—Prof. F. D. Richardson, Department of Metallurgy, Imperial College of Science and Technology, Prince Consort Road, London, S.W.7.

Lecturer in Physics at the University College of Rhodesia and Nyasaland—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1.

Lecturer (physicist, with experience and interest in high temperature gas physics) in the Department of Aerodynamics—The Recorder, The College of Aeronautics, Cranfield, Bletchley, Bucks.

National Research Council and other Post-Doctoral Fellowships, tenable in the Department of Physics—Prof. H. E. Duckworth, Chairman, Department of Physics, McMaster University, Hamilton, Ontario, Canada.

Scientific Officer (science graduate, preferably with honours in chemistry or biochemistry), for work on the preparation of blood products, including plasma protein fractions, with opportunity for research and developmental studies in an interesting and expanding field—The Director, Regional Blood Transfusion Centre, Royal Infirmary, Edinburgh 3.

Scientific/Senior Scientific Officers (with a first- or second-class honours degree in chemistry, or equivalent, and at least two years postgraduate research experience in organic chemistry, preferably with a bias towards the chemistry of natural products) at the Tropical Products Institute, D.S.I.R., London, W.C.1, to be members of small teams of research workers studying the isolation and structure of natural products possessing biological activity—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. F.849/9A.

Senior Lecturer in Pharmacology, and a Lecturer in Pharmaceutical Chemistry—The Registrar, Bradford College of Technology, Bradford 7.

Senior Cataloguer (chartered librarian, with experience in indexing scientific and technical information) in the College library—The Recorder, The College of Aeronautics, Cranfield, Bletchley, Bucks.

Senior Lecturer (qualified to direct and to teach to honours degree or Diploma of Technology levels, and preferably with industrial experience in a responsible position) in Textile Physics—The Registrar, Institute of Technology, Bradford 7.

Senior Physicist (with an honours degree in physics and experience in radiotherapy) with the Medical Research Council's Betatron Research Group, Christie Hospital and Holt Radium Institute, Manchester 20, to assist with the application of physics to the clinical use of the 4-MeV. Linear Accelerator and the 20-MeV. Betatron—Dr. Ralston Paterson, Christie Hospital and Holt Radium Institute, Manchester.

Technical Officer (man or woman, with an honours degree in mathematics, or in geography with a mathematical background) in the Survey Department, Government of Kenya, for survey computations and checking of survey plans—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.108/7/09.

Technician (Senior) (preferably with the Diploma of the Animal Technicians Association), to take charge of the Animal House—The Secretary, Guy's Hospital Medical School, London Bridge, S.E.1.

Technician (with a basic knowledge of, and an interest in, the maintenance and construction of electronic apparatus, and preferably experience in a physiological or pharmacological laboratory), to work in a new Department of Human Pharmacology—The Secretary, University College Hospital Medical School, University Street, London, W.C.1.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

Transactions of the Royal Society of Edinburgh. Vol. 63, Part 3 (No. 25). The Mineralogy and Metamorphism of the Moine Schists of the Morar and Knoydart Districts of Inverness-shire. By Dr. R. St. J. Lambert. Pp 553–588 12*s*. 6*d*. Vol. 63, Part 3 (No. 26): *Staurastrum paradoxum* Meye Meyen and *S. gracile* Ralfs in the British Freshwater Plankton, and a revision of the *S. anatinum*-group of Radiate Desmids By Dr. A J. Brook. Pp. 589–628 (18 plates). 17*s*. 6*d*. Vol. 63, Part 3 (No 27): A Structural History of the Girvan District, S W. Ayrshire. By Dr Alwyn Williams. Pp. 629–667. 21*s*. (Edinburgh. Royal Society of Edinburgh, 1959.) [1911

Commonwealth Relations Office: Colonial Office. Commonwealth Scholarship and Fellowship Plan: Proposed Arrangements for the Administration of the Plan in the United Kingdom. Pp. 5. (Cmnd 894). (London: H.M. Stationery Office, 1959.) 6*d*. net [2311

Other Countries

NORDITA (Nordisk Organisation for Teoretisk Atomfysik. Årsberetning 1957/58 (Annual Report). Pp. 29. (København: Nordisk Institut for Teoretisk Atomfysik, 1959.) [1611

The Undiscovered Earth: Proceedings of the Conference, June 11–12, 1959. Pp. 56. (Birmingham, Alabama: Southern Research Institute, 200 Ninth Avenue South, 1959.) [1611

Kungl. Sjofartsstyrelsen, Stockholm. Ergebnisse der Beobachtungen des Magnetischen Observatoriums zu lovo (Stockholm) in Jahre 1956 Von Folke Eleman und KjellBorg. Pp. 30 (Stockholm: Sjöfarsstyrelsen, 1959.) [1611

University College, Ibadan, Nigeria. Calendar, 1959–60. Pp 93. (Ibadan: University College, 1959.) [1911

Canada: Department of Northern Affairs and National Resources National Museum of Canada. Botanical Excursion to the Boreal Forest Region in Northern Quebec and Ontario. By W. K. W. Bladwin, E Lapage, J. Terasmae, D. W. MacLean and I. J Bassett Pp v+119 Botanical Excursion to Jasper and Banff National Parks, Alberta Alpine and Subalpine Flora. By A. E Porsild Pp ii+88 The Native Flora of Churchill, Manitoba, with Notes on the History, Geology, and Climate of the Area By H J. Scoggan. Pp. vi+51. Bulletin No. 160 (Biological Series No. 58). Contributions to Botany, 1958 Pp iii+148 (3 plates). Natural History Papers, No 3 (September 23, 1959) Anchiceratops from the Oldman Formation of Alberta. By W. Langston, Jr. Pp. 11. (Ottawa: Queen's Printer, 1959.) [1911

National Science Foundation, Washington Bibliography on the Economic and Social Implications of Scientific Research and Development. Pp. v+53. (NSF 59–41) 25 cents. Current Projects on Economic and other Impacts of Scientific Research and Development 1959. Pp. v+59. (NSF 59–51) 25 cents. (Washington, D.C.: Government Printing Office, 1959.) [1911

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DEVELOPMENT OF ATOMIC ENERGY IN BRITAIN

THE Atomic Energy Authority Bill, the second reading of which in the House of Commons on November 18 was moved by Sir David Eccles, amends two provisions in the Atomic Energy Authority Act, 1954. The first raises from ten to fifteen the maximum number of 'members' of the Authority, and the second enables the Authority to include in its pension scheme those of its staff who are transferred to the employment of the National Institute for Research in Nuclear Science. The reasons for these amendments were explained by Sir David and also, on December 3, by Lord Hailsham, when as Lord Privy Seal and Minister for Science he moved the second reading in the House of Lords.

Sir David Eccles said that the first clause of the Bill sought to recognize the growing complexity of the Authority's work and to restore a measure of flexibility for the future in the composition of the governing body while preserving the balance between full-time and part-time members. Since 1954, the staff of the Authority had increased from 17,000 to 37,000, and the Industrial Group alone now employs more than 18,000, a figure expected to exceed 23,000 by 1961–62. Between 1956 and 1959 the Authority increased its staff scientists and engineers by 71 per cent, and the figure of 4,220 is expected to increase by 42 per cent by 1962. At present, some 63 per cent are engaged on research and development, and the industrial consortia themselves are believed to employ some 4,000 men on reactor design and development.

The immediate requirements arise partly out of the recent decision to split the Industrial Group into a Development and Engineering Group, in charge of Sir William Cook, already a full-time member of the Authority, and a Production Group, of which Sir Leonard Owen had been designated head. It was desirable that Sir Leonard Cook, too, should be a full-time member. Further, it was desired to retain the services, as a part-time member, of Sir John Cockcroft, now Master of Churchill College, Cambridge, whose place as member for research had been taken by Sir William Penney. While at present the need is only for two further places, it was considered better to provide for a maximum of fifteen, rather than the twelve immediately required.

As regards the second clause, Sir David Eccles explained that the National Institute for Research in Nuclear Science, established to help the universities and other learned bodies participate in fundamental research in nuclear physics, is growing fast, and its staff is largely interchanged with the staff of the Authority. At present the Authority has no power to admit the staff of the Institute to its superannuation schemes if they so desire. Lord Hailsham made it clear that this permissive power would apply to both temporary and permanent exchange.

Although the Bill was generally welcomed in both Houses, there was considerable discussion on particular points in the House of Commons. Mr. A. Greenwood, who opened the debate, welcomed it as the first of any substance on the Authority since 1954; but while stressing the importance of part-time membership of the Authority as facilitating the participation in its work of distinguished men of science and industry, he expressed some misgivings as to consistency in the higher planning of the Authority's work. He recognized the need for flexibility, but although the Select Committee on Estimates had endorsed both the decision to split the Industrial Group and to restore executive responsibility to the technical members of the Authority in spite of the recommendations of the Fleck Committee, Mr. Greenwood said he is not entirely reassured. Stressing the importance of comparable conditions of employment in the Authority and at the National Institute for Research into Nuclear Science, he pointed out that in 1958 the Industrial Group had vacancies for 258 university graduates. The Authority had offered posts to 422 graduates, but only 160 accepted the appointments offered.

Mr. A. Albu, who also supported the Bill, urged the increase of Government expenditure on fundamental research as likely to benefit the industry, and referred to anxiety, particularly in the Isotopes Division, due to the frustration by Treasury control of the efforts of the scientists. Mr. Albu asserted that in the past two years many heads of departments in the Authority had resigned because of frustration, and he thought that this might bear on difficulties experienced in recruiting senior staff. The main administrative difficulty at present, he said, is lack of a sense of direction, and this is directly attributable to the retardation in the commercial development of reactors in consequence of the change in the energy position in Great Britain. Mr. Albu himself suggested concentration on a small type of reactor suitable both as a marine reactor and as a small land-based power station, which he thought should have a considerable market and assist and benefit the under-developed countries. He also thought that we should go much further in co-operating with other countries in Europe, but he did not mention that Britain's only opportunity with Euratom is now in the 1,000-megawatt nuclear programme which at the outside may be added to the 2,000-megawatts of nuclear plant which is being commissioned by 1965.

This plea for clearer understanding of the economics of nuclear energy was supported by Mr. H. Davies, who asked for a further opportunity for a constructive debate on nuclear energy and referred, in particular, to the need to release more information from the restrictions of secrecy. Another plea for a further debate came from Mr. R. Mason, who, questioning whether the decision to develop gas-cooled graphite-moderated reactors had been wise, expressed concern as to the shortage of scientists, technicians and engineers and, in particular, regarding the loss of so many highly qualified scientists abroad. He thought we should make a serious attempt to ascertain why so many good scientists, technologists and engineers, who accepted research scholarships in the United States and Canada, failed to return. The relations with industry also required consideration, and in particular we should find some way of using to a greater extent the consortia and their experienced staff.

In replying on the debate, Sir David Eccles assured the House that there is no intention of stopping work on the manufacture and sale of isotopes. The recommendations of the Select Committee on Estimates on the relations between the Authority and industry had been accepted by the Authority. Relations in reactor technology are already close, and the observations of the Committee on the manufacture of fuel elements are receiving attention. Referring to recruitment, Sir David said that the senior professional staff had increased from 377 on March 31, 1958, to 511 at the end of September; for scientists and engineers, the figures were 6,900 and 8,600, while the senior administrative staff had increased from 56 to 93. The Bill was passed without amendment at the Committee stage at the third reading on November 25.

The debates clearly lent much support to the findings of the Select Committee on Estimates in its Fifth Report for the Session 1958–59; but they should also stimulate that wider thinking about the development of nuclear energy in the changed context of high-level stocks of solid fuel, even if in Great Britain there are not the low fuel prices that, with large stocks, have caused Europe's need of nuclear energy to recede into the 1970's. The only way to sell nuclear power is manifestly to sell it cheap, and the Minister for Science should note the strong demand for regular debates on nuclear energy for the further reason that much constructive thinking is still needed on the relations between the Authority and industry and with Euratom. What is being done at Winfrith Heath will obviously be affected by the duplication and waste already apparent in the research programmes of the six member countries of Euratom. The new outlook for nuclear power inevitably forces on Great Britain the need for greater watchfulness to avoid waste of resources and man-power in its development, alike from the competitive point of view and from that of the efficient use of our own limited resources, with which the Minister for Science is intimately concerned. Meanwhile, it should be noted that, since the debate, it has been announced in London, on December 4, that teams of scientists from Great Britain and Euratom countries are to co-operate in research on controlled thermonuclear reactors, fast breeder reactors and advanced gas-cooled reactors. It is hoped that the new agreements, besides eliminating wasteful overlapping in research projects, may lead to more efficient use of scientific man-power, as well as to exchange of information, co-ordination in forward planning by the British and Euratom teams, and interchange of personnel between Britain and the Continent.

A RATIONALE FOR BEHAVIOUR

Biological and Biochemical Bases of Behavior

Edited by Harry F. Harlow and Clinton N. Woolsey. Pp. xx+476. (Madison, Wisc.: University of Wisconsin Press, 1958.) 8 dollars.

According to the preface of this book, the symposium which it represents derived from the activities of a sub-committee to explore the relationship between the biochemical and biological sciences and mental health, and its purpose was to correlate behaviour with results obtained from other biological sciences: anatomy, embryology, physiology, pharmacology and biochemistry. In the present state of knowledge, this might appear to be somewhat ambitious and the biochemistry of behaviour is, to say the least, a study for the future. Nevertheless, the rational basis of behaviour will be found only by the persistent study of the related basic sciences.

It is perhaps unfortunate that, for reasons of economy, the comments of those taking part and the general discussion have been omitted from the book, since it might be expected that this part of the symposium would produce more integration between different fields. As it is, the book consists of a number of isolated contributions, with only one common link; namely, that each represents some aspect of the study of behaviour. When compared with other similar publications of which there has been a spate in recent years, this book is perhaps unique in that the major contribution is provided by experimental psychology. Both the opening and closing chapters are written by psychologists (Harlow and Hebb) and both emphasize the contributions of psychological method to the study of behaviour. They also stress that the psychologist must turn to other disciplines for an explanation of the mechanisms underlying the phenomena he is studying, an important step which not all have taken. It is not surprising, therefore, that more than one-third of the book is concerned with the relation between function, in terms of behaviour, and structure; this is illustrated by the effects of lesions in the brain on various aspects of behaviour, for example, sensory discrimination. Such studies, examples of which are too numerous to quote, lend themselves most readily to the application of psychological methods.

Neurophysiological contributions to behaviour are represented by those of Magoun and Jasper, who discuss the brain stem reticular activating system in relation to sleep and wakefulness and the role of diffuse projection systems in integrative processes, such as conditioning. Lilly describes the plotting of a 'motor' map of the cerebral cortex from observations of movements produced by localized electrical stimulation utilizing 610 implanted electrodes! Olds deals with the relationship between electrical events and behaviour in self-stimulation studies, the full implications of which have yet to be clarified. There is one purely anatomical paper (Woolsey).

There are three biochemical contributions to this book. Dr. Tower effectively summarizes the present state of neurochemistry, emphasizing the effects of biochemical lesions, for example, phenylketonuria, on cerebral function. He concludes with remarks that the neurochemical substrates of cerebral activity must, at some stage, be incorporated into the neurophysiological and behavioural structure of the central nervous system. Rosenzweig, Krech and Bennett have, in fact, made an attempt to correlate neurochemistry and behaviour, and theirs is a rare study involving collaboration between biochemists and behavioural scientists. They have investigated the relationship between acetylcholine metabolism, measured by cholinesterase activity, and the maze behaviour of rats. This work is extremely interesting, and. if it does nothing else, it re-emphasizes the importance of biochemical studies on small areas of brain tissue. Indeed, any study which could throw light on the changes in the central nervous system accompanying the learning process is more than worthwhile. However, as Tower points out, not only is the role of acetylcholine in the brain equivocal but also cholinesterase activity is a doubtful measure of its metabolism.

Beach discusses sexual behaviour and its modification by hormonal and neuronal mechanisms. It is of interest to recall that profound changes in primitive behaviour of this type have been known to be affected by different chemical structures for some time. Recent advantages in the elucidation of the structure of hormones and their chemical synthesis should provide an even sounder base-line for future study.

A book of this nature usually provides a fund of vogue words; "interdisciplinary" and "major breakthrough" have been met before but "ideational" is new to us. As is inevitable in publications of this nature, much of the material has been published elsewhere, either before or since the symposium was held, while other contributions represent entirely new work. This does not detract from the value of the book nor does the fact that it is now three years since the symposium took place. It is a useful guide to current thought and knowledge and should be read by all interested in the study of behaviour. The book is provided with an adequate subject index.

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CRYSTALLOGRAPHER'S TOOL-CHEST OF MATHEMATICS

International Tables for X-ray Crystallography
Edited by John S. Kasper and Kathleen Lonsdale. Vol. 2: Mathematical Tables. Pp. xviii+444. (Birmingham: The Kynoch Press, 1959.) 115*s*.

THESE tables form the second instalment of the complete revision and expansion of the original "Internationale Tabellen" of 1935. The history of these tables and of the present project have been described by Ewald in his review (Ewald, P. P., *Nature*, 171, 944; 1953) of Vol. 1 of the present tables. Vol. 2 has been compiled with the object of including all the formulæ and mathematical tables that a crystallographer may be reasonably expected to require. They contain no data that depend on physical measurement; such data will appear in Vol. 3.

Perhaps the most significant innovation in these tables is the inclusion of a 90-page section on mathematical methods. This covers algebra, trigonometry (plane and spherical), calculus (integral and differential), vector and tensor analysis, Fourier-transform theory and statistics, and comprises definitions, theorems, methods of attack, and examples of practical applications to crystal structure analysis. Here one can find readily, for example, how the characteristic values and vectors of a matrix may be calculated; what methods are available for the numerical solution of transcendental and other 'insoluble' equations, and what are their relative merits and particular applications; what are the general properties of groups; how the Fourier transform of a discontinuous function may be expressed in terms of the discontinuities; how to apply a statistical significance test; over what range of values of x is $\sin x$ equal to $x - 1/6\, x^3$ to within 0·001; how a given number may be approximated to a rational fraction by the use of continued fractions. There are many references to standards texts and to special articles. It is surprising, however, that interpolation formulæ are not treated, since these should be within easy reach of anyone who uses tables, and are moreover useful whenever the question of the most suitable interval for the tabulation of mathematical functions or of experimental observations is under consideration.

Under the heading "Crystal Geometry" there are definitions and formulæ relating to direct and reciprocal lattices, angles between planes, twinning conditions, and quadratic forms. A section on diffraction geometry deals with all the well-known techniques for recording X-ray diffraction patterns; their particular characteristics and applications are listed and formulæ and tables relevant to each method are given. The accuracy of measurement of lattice parameters is treated in considerable detail. "The Physics of Diffraction Method" covers formulæ for the intensity of diffraction by a crystal, and the corrections for temperature effects, absorption and extinction. Under "Fourier Synthesis and Structure Factors" appear formulæ for the various types of synthesis that are in use, and methods of structure refinement are fully treated. There is also a survey of digital and analogue methods of numerical Fourier synthesis.

Further topics are the close packing of spheres in various space groups, statistical tests on intensity data for the detection of symmetry elements, and inequalities between structure factors that may give information about their phases. (The *probable* relations between phases are not included, however.) Finally, there are some useful sets of trigonometrical tables (tailored for crystallographers), and a glossary of crystallographic terms in English, French, German, Spanish and Russian, in rather more detail than that in Vol. 1.

The criteria on which such tables are to be judged are those of comprehensiveness, ease of use, and comprehensibility. On the first and second scores it is almost impossible to find any fault. The compilers took the view that nothing of potential application should be left out, since few crystallographers have a perfect memory for formulæ, and most prefer not to spend time working them out or searching textbooks. A very full and systematic contents list makes all material easily accessible, and renders an alphabetical index unnecessary. On the third score it is again difficult to find fault, but one wonders whether perhaps mathematical elegance and logical development have occasionally been given first place rather than ease of reference; for example, the introduction to the section on "Fourier Theory" (surely a slight misnomer because Fourier also had a theory of heat conduction) might well scare the weaker mathematician into thinking that he has come to the wrong book. But this criticism does not apply generally.

The layout and printing are of excellent quality, and the binding is pleasing in appearance and made for hard use. The price is in line with that of any first-class text-book of to-day; if it saves a laboratory a total of five days of research-worker time or one hour of computer time it will have yielded a handsome profit.

A. R. STOKES

THOUGHTS ON COMMUNICATION

The British Association Granada Lectures

Science and Communication. By Sir Edward Appleton. Television and Politics. By Edward R. Murrow. Dons or Crooners? By Sir Eric Ashby. (Three Lectures given in Guildhall, London, in October 1959, on the subject of "Communication in the Modern World".) Pp. 111. (London: Granada TV Network, Ltd., 1959.) 3*s*. 6*d*.

THESE three lectures were organized by the British Association and sponsored by Granada TV Network, Ltd. They were delivered in Guildhall, telecast later in a considerably shortened form, and now appear in print. This immediately presents one with the problem of how to treat them: as lectures, television or essays? Not having seen them on television, I will ignore this aspect.

The general title of the three lectures is "Communication in the Modern World". None of the lectures conflicts with this title, but they are related only formally.

The first lecture, by Sir Edward Appleton on "Science and Communication", gives a lucid account of modern communications, but says little that is new. One expects a few original ideas in a 'prestige' lecture of this sort. He might well have extended the section where he speculated about future developments. From a man so eminent in the field of communications, this would have been far more interesting than a summary of our present knowledge, as already several good ones exist.

Dr. Edward R. Murrow gave the second lecture on "Television and Politics". He throws out ideas, wittily recounts numerous telepolitical anecdotes and very wisely draws the final conclusion: "In the area of political television we are all ignorant". Another point he makes, rather more tentatively, is that it is personalities that come across, not ideas. For example, Vice-President Nixon during his election campaign was accused of accepting too many presents. He appeared on television and won overwhelming support from the public by refusing to give up the one present he admitted to receiving—a little dog—because "the kids love that dog". Similarly, Dr. Murrow ascribes McCarthy's rise largely to his flair for publicity on television, and he suggests that Stevenson would have done better in a non-television election where he was not up against Eisenhower's homely personality. But—"It is a limited medium . . . it can arouse curiosity . . . entertain . . . sell goods. But it can't reason . . ."

Sir Eric Ashby, in his lecture "Dons or Crooners?", is the only one really to put forward a thesis. It is this: The ordinary man does not feel that he really belongs to society, as he did, say, in the Middle Ages. This is because he is unable to understand the science on which it is based. Therefore, he must be given some understanding of science, and this should be done by emphasizing the importance of technique in science, for the ordinary man is a craftsman and he appreciates craftsmanship. This is an excellent plan so long as it is not regarded as the only one. But Sir Eric is very patronizing about the ordinary man. He is called Ron Blossom and he lives in Doris Grove, Birmingham. His response to a dramatic scientific discovery is to say "Cripes". One thing no one can afford to do if he wants to put an idea across without antagonizing the recipient is to patronize him. Surprisingly for the author of "Technology and the Academics", Sir Eric says: "The interpretation of science to the likes of you is no problem. You have the Third Programme and Pelicans . . ."; 'you' in this case being a 'Third Programme audience'. Now it is only true that the interpretation of science to highly intelligent people presents no problems if the people are interested. The trouble is that a great many of them are not. Where there is a will there may be a way. But it does not follow that where there is a way there is a will.

B. SILCOCK

DESCRIPTIVE ASTRONOMY

Elementary Astronomy

By Prof. Otto Struve, Beverly Lynds and Helen Pillans. Pp. vii+396. (London and New York: Oxford University Press, 1959.) 55*s*. net.

THIS is a text-book based on a new approach to the study of astronomy. Many elementary texts have considered primarily the historical and purely factual descriptive aspects of astronomy. Recently, there has been felt a growing need for a course emphasizing the physical basis of astronomy, using at most the simplest mathematics to perform calculations illustrative of the physical principles involved, but still including enough descriptive material to give the student a basic knowledge of modern astronomy. This book aims at meeting this need, and it fulfils its intentions admirably. The subjects are treated in more or less conventional order, starting with the Earth, Moon and planets, followed by the Sun, the stars and star clusters, and ending (apart from a chapter on instruments) with galaxies, but the book has quite a different tone compared to older texts. Rarely has a volume intended for serious students been so profusely illustrated, excelling most of the avowedly popular books. Still more rarely has any general text-book contained so many illustrations from the recent periodical literature. The senior author has in the past attained a reputation for his descriptive articles no less than for his research work, and this book fully maintains this reputation.

Astronomy, in common with all other sciences, is developing very rapidly, and no one can be an expert in all its branches. Yet the whole of this book is surprisingly up to date, a feature which will commend it to teachers of astronomy. Most of the book will be intelligible to amateur astronomers, and it should find a place on the shelves of every general library with an astronomy section. The authors state in the preface that their book is not intended to replace the many excellent text-books of descriptive astronomy now in use. The reviewer is confident that it will indeed become the leading introductory text on its subject.

R. H. GARSTANG

Recent Advances in Atmospheric Electricity

Proceedings of the Second Conference on Atmospheric Electricity, held at Portsmouth, New Hampshire, May 20–23, 1958. Edited by L. G. Smith. (Sponsored by Aerophysics Laboratory, Geophysics Research Directorate, Air Force Cambridge Research Center, Air Research and Development Command.) Pp. xv+631. (London and New York: Pergamon Press, 1958.) 120*s.* net.

THIS book reproduces the fifty or so papers that were presented at the conference named in the sub-title. They are arranged under three sub-divisions: fair-weather electricity, thunderstorm electricity and the lightning discharge. In some cases a résumé of the discussions following a paper is included. There does not appear to have been any attempt at making a selection of the more important contributions for publication and in consequence the value of individual papers varies widely. It is difficult to find out from such a varied assortment of material how much of it does really represent recent advances in the subject; some of the papers describe work that is already well known, while others are more in the nature of interim reports.

It is significant that a large part of the work described is devoted to atmospheric electrical investigations in the upper air by means of aircraft measurements and radio sondes. Another significant trend is the use of radar in studying thunderstorm precipitation and lightning.

Interested people who were unable to attend the conference will welcome the opportunity of learning what went on there, and there is no doubt that the active worker in the subject can find a great deal of interest to him in many of the papers reproduced in the book. But it is very doubtful whether a book as expensive as this, beautifully produced though it is. will attract many individual purchasers.

F. J. Scrase

Principles of Organic Chemistry

By T. A. Geissman. Pp. viii+635. (San Francisco: W. H. Freeman and Company; London: Bailey Bros. and Swinfen, Ltd., 1959.) 7 dollars; 56*s.* 6*d.*

THE author has written an elementary text-book of organic chemistry which includes most of the material required by a first-year university student, but which has its emphasis on important functional groups and on general types of organic reactions. This approach leads to obvious omissions in a book of such moderate size, but Prof. Geissman holds very reasonably that the student's time is initially better spent in the study of principles than in the acquisition of general information. The last sixty pages, however, are largely descriptive and deal with structurally more complicated molecules, many of which are biologically important. Thus the discussion of the basic principles of the subject has been complemented by some of its major achievements and applications. In this way, the student is shown some of the scope of organic chemistry.

Prof. Geissman has, on the whole, succeeded in his objects. The presentation and printing of the book are both good, except that some of the diagrams of molecular models are confusing and that a few errors in formulæ may be misleading to students. Perhaps the allocation of space to various topics is a little unbalanced. For example, more room is given to aromatic electrophilic substitution than to stereochemistry. Nevertheless, this book can be enthusiastically recommended as an interesting and stimulating introduction to organic chemistry.

C. B. Reese

Introducing Astronomy

By J. B. Sidgwick. Pp. 259. (London: Faber and Faber, Ltd., 1959.) 6*s.* net, paper bound.

THIS book is a fairly drastic revision of the same author's "Astronomy for Night Watchers" and, as such, is aimed at promoting an interest in astronomy in the layman; no knowledge of mathematics or physics being presumed. It is divided into two roughly equal parts. After two introductory chapters, the first part consists of an elementary outline of descriptive astronomy, taking the usual route outwards from the Sun to extra-galactic nebulæ; followed by simple explanations of some astronomical instruments. The second part is almost entirely taken up with a description and sketch of each constellation in turn, mentioning such objects of especial interest as may be observed with binoculars. These artificial divisions may be a disadvantage from an observer's point of view but allow the inclusion of a brief history and mythology of the constellations.

Mr. Sidgwick writes with an easy style and has succeeded in producing a very readable and balanced introduction to astronomy with very few errors, none of which seriously detracts from its value. Definitions are introduced as necessary and there are numerous cross-references, so the beginner should encounter few, if any, stumbling-blocks. The reader whose new interest in this old science has been so ably fostered is likely to want to know more. It seems a pity, therefore, that space has not been spared for even a short bibliography.

B. R. Leaton

The Birds of the British Isles

By Dr. David Armitage Bannerman. Vol. 8: Phalacrocoracidae, Sulidae, Fregatidae, Procellariidae, Diomedeidae, Podicipedidae, Gaviidae, Columbidae, Pteroclididae. Pp. x+400+26 plates. (Edinburgh and London: Oliver and Boyd, Ltd., 1959.) 63*s.* net.

THE eighth of Dr. Bannerman's impressive volumes is largely concerned with sea-birds—the cormorants, the gannet and the petrels. The magnificent frigate-bird is included on the strength of its sole recorded appearance, in 1953, and the black-browed albatross in view of two certain and a few doubtful records; with these, as also with the rarer petrels, the author follows his practice of giving full information from the regions where the birds more truly belong. Then come the grebes, the divers, the pigeons and, finally, Pallas's sandgrouse. The remarkable spread of the collared turtle-dove, unknown in Britain until the present decade, is duly chronicled; so also are the extraordinary occasional visitations of the sandgrouse from the Aral-Caspian region. To an increasing extent Dr. Bannerman has included contributions from other writers. Thus the whole chapter on the gannet is by Mr. George Waterston and those on two of the exotic petrels are by Dr. Alexander Wetmore, while various other chapters have sections by ornithologists with special knowledge of particular aspects.

Landsborough Thomson

MAGNETOTROPISM: A NEW PLANT-GROWTH RESPONSE

By Prof. L. J. AUDUS
Botany Department, Bedford College, London

TWO major theories of graviperception now stand opposed. One due to Brauner[1] is based on the so-called geoelectric effect. Apart from the uncertainty cast on this whole phenomenon by recent investigations[2], I have long felt that the forces involved were utterly inadequate to cause the extremely rapid and extensive changes in hormone concentration required by the observed rates of growth response. In the second and classical statolith theory, cell particles having a density different from that of the surrounding cytoplasm move under gravity to one side of the responding cell, thereby evoking a growth response. Starch grains in certain cells of virtually all receptor organs studied respond in this way by sinking to the lower side of the cell[3]. How such lateral migration of starch grains could cause rapid changes in hormone distribution has already been discussed elsewhere[4].

The fact that organs respond to laterally applied electrostatic potential gradients has been regarded as very strong support for the geoelectric theory, although recent findings[2] call for a major revision of ideas on this phenomenon. No similar kind of support has been forthcoming for the starch-grain statoliths because so far no forces other than those arising from the mass of the grain itself (that is, gravity, centrifugal force) have been employed to shift them in the cell. In consideration of the possibilities of circumventing this difficulty, attention was given to the behaviour of organic substances in strong magnetic fields. They are normally weakly diamagnetic, and if therefore particles of such materials are placed in a *non-uniform* magnetic field, then a force will be exerted on such particles in the direction of the maximum gradient and equal to:

$$(K_1 - K_2)v.H.\frac{dH}{dx}$$

where K_1 and K_2 are the volume susceptibilities of the particle and suspending medium respectively, v is the volume of the particle, H the magnetic field strength and dH/dx the field gradient.

Few, if any, experimental observations have been made on the magnetic susceptibilities of starch or protein solution, like that surrounding statolith starch in the cytoplasm of the plant cell. Since exact experimental determination of these unknown parameters promised to involve much time and the employment of special techniques, it was resolved, in the first place, to study the behaviour of a suitable organ in a steep magnetic gradient. To this end a large permanent magnet, designed for use with a magnetron, was employed. This was fitted with soft iron pole pieces of triangular cross section as in Fig. 1. Direct measurements of the field strength by the Quincke method showed that such an arrangement gives maximum field strength just inside the narrowest point of the gap and the steepest gradient in an outward direction in the plane A–B, as is shown by the graph of Fig. 1. The field strength in this plane was 4,000 gauss and the gradient—5,600 gauss per cm.

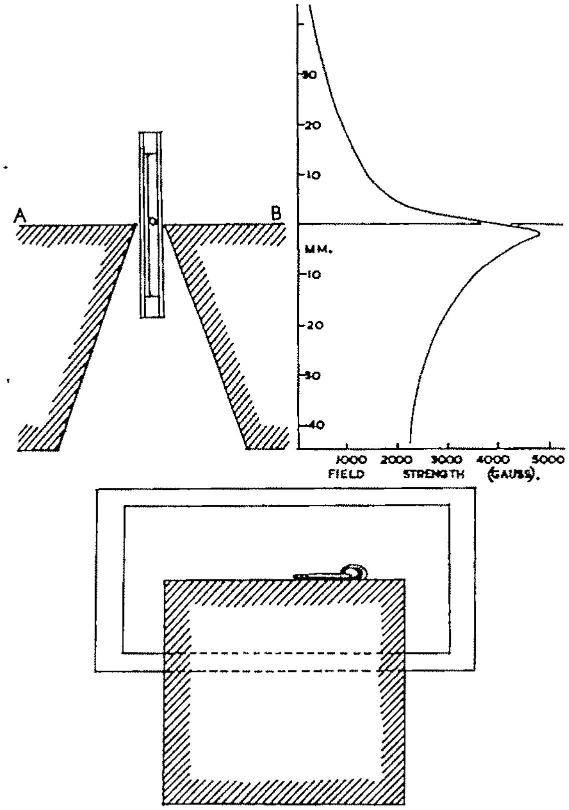

Fig. 1. Diagrams showing magnet gap design, position of glass growth chamber and cress seedling roots and field-strength distribution through the gap. Top, cross-section and field-strength curve; bottom, end view

The organ chosen for study was the seedling root of *Lepidium sativum* growing under sterile conditions on plain 2 per cent agar in a small glass chamber of design shown in Fig. 1. The seedlings were germ-

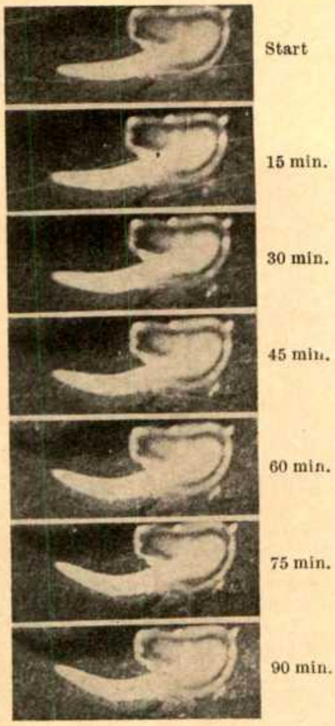

Fig. 2. Progress of curvature of 36-hr. roots of *Lepidium sativum* seedlings down the magnetic gradient

inated and grown for 36–60 hr. in the dark in an incubator at 25° C. and the roots allowed to grow downwards along the vertical surface of the agar. At the commencement of the treatment the chamber was inserted as illustrated between the poles of the magnet so that a root lay parallel to the gap and slightly above it, as near as possible to the region of maximum force. The magnet, which was held in a heavy brass cradle and mounted on an axle in ball bearings was rotated about the axis of the gap at a speed of about 2 revolutions per min. to eliminate gravitational effects which might swamp any magnetic effects. At intervals of 10–15 min. the root tip and edge of the nearest pole piece were photographed from the direction $A \rightarrow B$ by an automatic 35 mm. Robot camera provided with an 8-in. lens giving an image almost natural size on the film. The photographs were taken with synchronized electronic flash illumination of 1 µsec. duration without stopping the rotation. The direction of flash illumination was thus at right angles to the magnetic gradient; but, in any event, as shown by experiments employing visual observation only, this flash illumination had no detectable effects either on control roots out of the field or on those situated just above the gap. All these observations were made at room temperatures (18–22° C.) and in weak diffuse light.

A large number of experiments have now been performed, and in all but a very few cases roots have shown marked growth curvatures away from the gap, that is, down the magnetic gradient. Typical results for a 36-hr.-old root and 60-hr. roots are shown in Figs. 2 and 3 respectively. From the photographic records, preliminary estimates have been made of the reaction time by extrapolating backwards the curvature progress curve to cut the axis of zero curvature. In the fluctuating summer temperatures of the laboratory (18–22° C.) the values obtained fell between 30 and 45 min. Under similar environmental conditions measurements have shown the time of reaction to a continuously applied gravitation stimulus to be 4–8 min.

Having obtained such success with growth responses, attention was then focused on starch grains. First, the value of their diamagnetic susceptibilities was considered. By use of the Pascal constants it is possible to calculate from their molecular structures the magnetic susceptibilities of organic molecules. Assuming for these purposes starch grains to be amorphous the average volume susceptibility (K_{ST}) can be estimated to fall between about $-0\cdot74$ and $-0\cdot62 \times 10^{-6}$ c.g.s. units depending on the value of the density of the starch grain taken to derive it from the specific susceptibility χ. The average value of volume susceptibility for the surrounding cytoplasm (K_P) is much more difficult to assess but should differ very little from that of water, that is, $-0\cdot72 \times 10^{-6}$ c.g.s. units. The difference in the two susceptibilities will therefore fall between $+0\cdot10 \times 10^{-6}$ and $-0\cdot02 \times 10^{-6}$ and the resultant force on the grain may be either inwards towards the gap or outwards away from it, depending on the particular value of K_{ST}. However, starch grains are not amorphous and it is therefore possible, as has been shown for cellulose[5], that they may show appreciable diamagnetic anisotropy. If we make the reasonable assumption that the maximum value of K_{ST} along a particular axis of the grain could rise to 25 per cent above the average susceptibility calculated above, then this maximum value of K_{ST} falls between $-0\cdot93$ and $-0\cdot78 \times 10^{-6}$ c.g.s. Since the starch grain would orientate itself in the field so that its axis of maximum susceptibility would be perpendicular to the lines of magnetic flux, that is, along the maximum field gradient, then the effective force on the grain would consistently be away from the gap. It is a simple matter to get an estimate of this magnetic force in terms of the gravitational

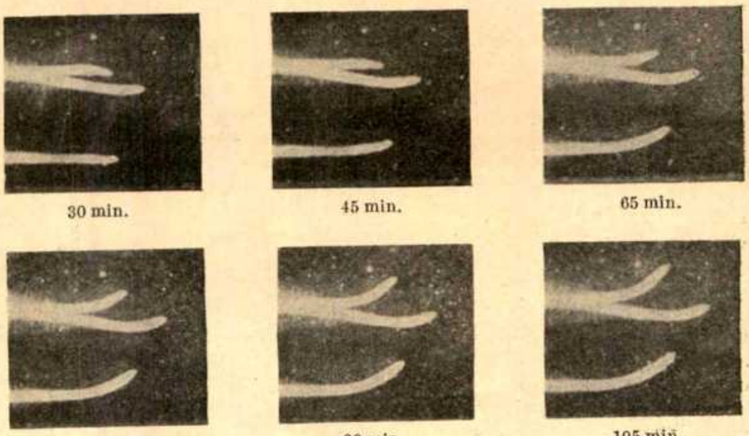
30 min. 45 min. 65 min.
75 min. 90 min. 105 min.

Fig. 3. Progress of curvature of 60-hr. roots of *Lepidium sativum* seedlings down the magnetic gradient

force acting on the same grain. Thus the gravitational force is expressed by the formula:

$$(\rho_{ST} - \rho_P)v.g$$

where ρ_{ST} and ρ_P are the densities of starch and protoplasm respectively. The ratio of the magnetic force to this force is given by the formula:

$$\frac{(K_{ST} - K_P)}{(\rho_{ST} - \rho_P)} \cdot \frac{H}{g} \cdot \frac{\mathrm{d}H}{\mathrm{d}x}$$

The value of ρ_{ST} can be estimated only approximately. Dry potato starch has a density of 1·5, which falls to 1·25 when the grain is fully imbibed. Egg white, taken as near model of cell protoplasm, gives an estimate of $\rho_P = 1{\cdot}04$. Depending then on the density taken for the statolith starch grain we obtain numerical values for this ratio ranging between 0·01 and 0·0065. This ratio is very small compared with the ratio of reaction times for magnetotropic and geotropic curvatures respectively. It is possible, however, that the ratios of presentation times, which would presumably be directly related to starch grain movement, might more nearly approach this calculated ratio of available forces.

With these possibilities in mind, attempts have been made to observe the behaviour of starch grains in roots bending in this magnetic field. Thus, with the minimum of delay after rotation was stopped, curved roots were removed from the experimental chamber and fixed in formalin–acetic–alcohol. They were first placed on the surface of a small disk of radish tissue in a film of sodium alginate dissolved in diluted indian ink, and carefully and quickly orientated thereon so that later the plane of cutting in the paraffin block could be easily and accurately identified. The whole was flooded with fixative containing 1 per cent calcium chloride simultaneously to fix the root and to turn the sodium alginate solution into a rigid calcium alginate gel. After embedding in paraffin wax, longitudinal sections were cut at 8μ in the plane of bending and stained for 24–48 hr. in dilute aqueous iodine. Measurements of starch grain distribution were made on all the root-cap cells appearing to be complete in one or more median or near median sections of well-curved roots. By using an ocular with a median hair line, which was adjusted to bisect the cell as nearly as could be judged by eye in a direction parallel to the main root axis at the tip, the numbers of starch grains on the two sides of each cell were counted and differences determined. Results of these preliminary observations are shown in Table 1. It will be seen that in most sections there is a very slight preponderance of grains in that half of the cell nearer the concave side of the root, that is, the side away from the magnet. In two sections only is this shift significant at the 5 per cent level and in most cases it is not significant. However, the overall picture is undoubtedly one of a slight shift of starch grains down the magnetic field gradient.

Suggestive as they are, these preliminary experiments do not yet constitute proof of the statolith starch theory of graviperception. They must be followed by more precise studies on starch grain behaviour in root caps and with completely unambiguous and objective methods in order to eliminate any possibility that the very small shifts recorded are not artefacts of technique or observation. In addition, exact experimental determination will have to be made of the magnetic properties of these starch grains both inside and outside the cell and of the surrounding protoplasm if suitable methods can be found for studying them. The present experiments do most strongly suggest, however, the involvement of some moving material which distributes itself in the cell under a magnetic gradient in the same sense as it does under gravity. Thus if it were a cytoplasmic particle it would have to be either denser than cytoplasm with a higher diamagnetic susceptibility, or less dense with a smaller susceptibility. However, we must not lose sight of the fact that the root as a whole will be subjected to a small outwardly directed force which will set up small bending stresses in the growing zone. The part such stresses may play in the growth responses observed must also be fully elucidated.

Table 1

Root	Section (numbered in series)	Average number of starch grains per cell	Average difference in number of starch grains. Concave–convex side	$\sigma/\sqrt{N}$	N	P
1	Median	7·40	0·79	0·37	33	0·05
2	Median	5·75	0·58	0·48	12	n.s.
3	1	5·17	0·27	0·66	11	n.s.
	2	6·1	0·5	0·63	10	n.s.
	3	6·95	1·50	0·62	20	0·03
	4	6·53	0·75	0·39	28	n.s.
	Median	6·55	0·27	0·36	29	n.s.
	Median	5·23	0·32	0·36	38	n.s.
	7	6·08	0·35	0·35	37	n.s.
	8	7·04	1·0	0·56	22	n.s.
	9	6·46	0·36	0·35	28	n.s.
	10	9·00	1·29	0·54	21	0·03
	11	8·27	−0·61	0·48	18	n.s.
	12	9·21	0·21	0·98	14	n.s.
4	1	7·43	−0·48	0·47	23	n.s.
	2	7·64	0·91	0·81	11	n.s.
	Median	8·17	0·0	0·53	24	n.s.
	3	11·01	0·17	0·73	12	n.s.
5	1	7·92	1·32	0·62	28	0·05
	2	6·00	0·3	0·36	20	n.s.
	Median	7·55	0·64	0·51	22	n.s.
	4	7·20	0·09	0·80	11	n.s.

So far the phenomenon has been investigated with only one organ of one plant species. It must be extended to roots of other species and also to other organs, such as the coleoptile, to see how universal is the correlation between the responses to gravity and those to a magnetic gradient.

Such experiments are being actively pursued, and it is hoped that this new phenomenon may prove to be a valuable tool in our attack on the problem of the mechanism of graviperception.

I wish to acknowledge my indebtedness to Dr. V. I. Little of the Physics Department, Bedford College, for many valuable discussions, for advice in the design of apparatus and for arranging the loan of the magnet used; to Dr. V. C. G. Trew of the Chemistry Department, for information on magnetochemistry and the calculation of diamagnetic susceptibility of starch, and to Dr. B. C. Sharman of the Botany Department, Bedford College, for suggesting the use of calcium alginate as a cement in the orientation of roots for embedding.

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[1] Brauner, L., *Jahrb. wiss. Bot.*, **66**, 781 (1927); **68**, 711 (1928).

[2] Brauner, L., *Planta*, **53**, 449 (1959).

[3] Rawitscher, F., "Der Geotropismus der Pflanzen", 308 (Gustav Fischer, Jena, 1932).

[4] Audus, L. J., and Brownbridge, M. E., *J. Exp. Bot.*, **8**, 235 (1957).

[5] Selwood, P. W., "Magnetochemistry" (2nd edition), 127 (Interscience Pub., Inc., London, New York, 1956).

RADIATION CARCINOGENESIS: A NEW HYPOTHESIS

By Dr. P. R. J. BURCH

Medical Research Council Environmental Radiation Research Group, Department of Medical Physics, University of Leeds, The General Infirmary, Leeds 1

THERE is growing scientific and social concern about the possible influence of small doses of ionizing radiation upon the incidence of leukæmia and bone cancer in human beings. That large doses of X-rays (of the order of 1,000 rems) can occasionally induce leukæmia, and that long-standing radium burdens (of the order of several μgm.) in the human skeleton can occasionally produce bone cancer, are facts which are not in dispute. However, at doses and dose-rates comparable to background (generally about 0·1 rem/yr.), changes in cancer incidence are likely to escape detection because on any plausible theory they will represent only a small fraction of the normal incidence. To predict the somatic effects of small amounts of radiation we are therefore compelled to use statistics relating to high doses, and to extrapolate to low by means of a theoretical dose–response relationship.

The linear theory of carcinogenesis, which states that the incidence of radiogenic cancer is directly proportional to the radiation dose, has recently been criticized so effectively by Brues[1] that it must be regarded as being of very doubtful validity. Kimball[2] challenges the reliability of the statistics and statistical tests used by Lewis[3] in support of a linear theory of radiation leukæmogenesis. Mewissen[4], analysing the dose–response relationship in radiation leukæmia by the method of marking probits, suggests that leukæmogenesis may require more than one critical molecular event, although a single event may be the initiating agent.

Experiments in which osteogenic sarcomas in mice were induced by bone-seeking radioactive isotopes are regarded by Finkel[5] as giving evidence for a threshold effect. The low doses received by human beings from world-wide radioactive fall-out will not, she concludes, induce a single osteogenic sarcoma. By contrast, a maximum upper limit is predicted[6] from the linear theory of nearly ten cases per annum per million of population (for strontium-90 at an equilibrium-level in bone, assuming continuation of bomb tests). In view of the great diversity in these estimates, it is necessary to re-assess the evidence and to consider whether there is any alternative to the linear and threshold theories.

In this article, human radiogenic leukæmia and bone cancer will be considered, and arguments will be advanced in support of the tentative view that both diseases are initiated by a highly specific 'two-hit' mechanism.

Dose–Response Relationship in Human Leukæmogenesis

Court Brown and Doll[7] examined the statistics for the incidence of leukæmia in patients irradiated for ankylosing spondylitis and adopted the working hypothesis that there is no threshold dose, and that at low doses the incidence of radiogenic leukæmia is proportional to dose. Their results are re-plotted on log–log scales in Figs. 1 and 2. In Fig. 1, where the standardized incidence in excess of normal (that is, the radiogenic incidence) is plotted against the mean dose to the spinal marrow, it appears that a line of slope 2 gives at least as good a fit to the points as a line of slope 1. This implies that the radiogenic leukæmia incidence-rate might equally well be proportional to the square of the mean dose to the spinal marrow as to the first power. Court Brown and Doll refer, however, to the unreliability of the point at highest dose where extra-spinal marrow received a larger proportion of the dose than at the lower dose-ranges.

In Fig. 2, the radiogenic incidence-rate is plotted against the whole-body integral dose. Since the statistical errors will be similar to those in Fig. 1, the fit of the points to a line of slope 2 is very good, and it suggests that the radiogenic incidence-rate might be proportional to the square of the whole body integral dose. These quadratic relationships have been briefly noted previously[1,8], and they hint at the possibility that radiogenic leukæmia may require a two-hit process for its initiation. It will be seen later, however, that if the hypothesis elaborated here is true, this evidence will require very extensive re-interpretation before valid conclusions can be drawn from it concerning the dose–incidence relationship.

Consideration of other evidence of human radiogenic leukæmia will be deferred until after the mathematical relationships have been presented.

Dose–Response Relationship in Human Bone Cancer

The scanty data[9] for mortality from bone cancer as a function of terminal radium burden are plotted on log–log scales in Fig. 3. They imply that radiogenic incidence is proportional to the square of the terminal radium burden. Differences in radium uptake will give rise to different deposition patterns in bone. Despite this complication, for long-term burdens there may be a rough correspondence between the square of the average terminal burden for a group and the effective mean square dose to relevant bone tissue. If this can be assumed, then for radiogenic bone cancer, as for leukæmia, there is a suggestion that a two-hit process is involved.

The formal properties of two contrasting two-hit models will now be considered.

Fisher's Multiple Mutation Theory

The idea that natural or spontaneous cancers may arise through a succession of somatic mutations has been put forward on many occasions[10], and most recently by Fisher[11], who postulates the following sequence of events: (1) a somatic mutation; (2) clone growth from the mutated cell; (3) a second mutation within a clone cell which, in the case of a two-mutation sequence, will give rise to cancerous growth.

In the case of a three-mutation sequence, (3) above is followed by: (4), a second clone-growth of cells containing two mutations, and (5), a third mutation within this clone leading to tumour development.

Fisher also argues that in epithelial tissues it is likely that the number of mutated cells in a clone increases (after the first few divisions) in proportion to the square of the time. If it is considered that a *natural* cancer arises as a result of n successive spontaneous mutations, and if mutation-rates and clone growth-rates are invariant with respect to age, the age-specific incidence-rate is shown by Fisher to be proportional to $(\text{age})^{3(n-1)}$. Thus, for $n = 2$, the age-specific incidence-rate would be proportional to $(\text{age})^3$ and for $n = 3$ it would be proportional to $(\text{age})^6$. Court Brown and Doll[12] have examined the natural death-rate versus age relationships for chronic lymphatic, chronic myeloid and acute leukæmias. With chronic lymphatic leukæmia there is probably no genuine cohort effect[12] (that is to say, the actual death-rate from chronic lymphatic leukæmia at a given age is not changing with time) and the age-specific death-rate is proportional to $(\text{age})^6$; thus, assuming the difference between age at incidence and age at death will not appreciably distort the relationship, a triple-mutation sequence would be implied. For chronic myeloid leukæmia it is again likely that there is no genuine cohort effect[12], and for this disease the age-specific death-rate is proportional to $(\text{age})^3$, suggesting a double-mutation sequence. Acute leukæmia shows a marked cohort effect between the years 1945 and 1957 and the shape of the age-specific death-rate versus age curve is unusual, with mortality falling from adolescence to a minimum at 25–29 years, and then increasing slowly up to 70 years. Since the cohort effect suggests the presence of an extrinsic leukæmogenic agent, no reliable conclusion can be drawn about a possible intrinsic death-rate versus age relationship, except that from Court Brown and Doll's graph it appears unlikely to be of the sixth-power type.

It may well be significant that radiation exposure has been associated with chronic myeloid and acute leukæmias, but not with chronic lymphatic leukæmia requiring (hypothetically) a three-hit rather than a two-hit sequence.

Equally important is the connexion between age-specific death-rates from bone cancer and age. It is possible that there is no genuine cohort effect where incidence (as opposed to death) is concerned[13], and plotting from Case[13] the mean age-specific death-rate from cancer of bones (excluding jaw) for the 1866 and 1871 cohorts against age on log–log scales, we find, from Fig. 4, dependence on $(\text{age})^3$.

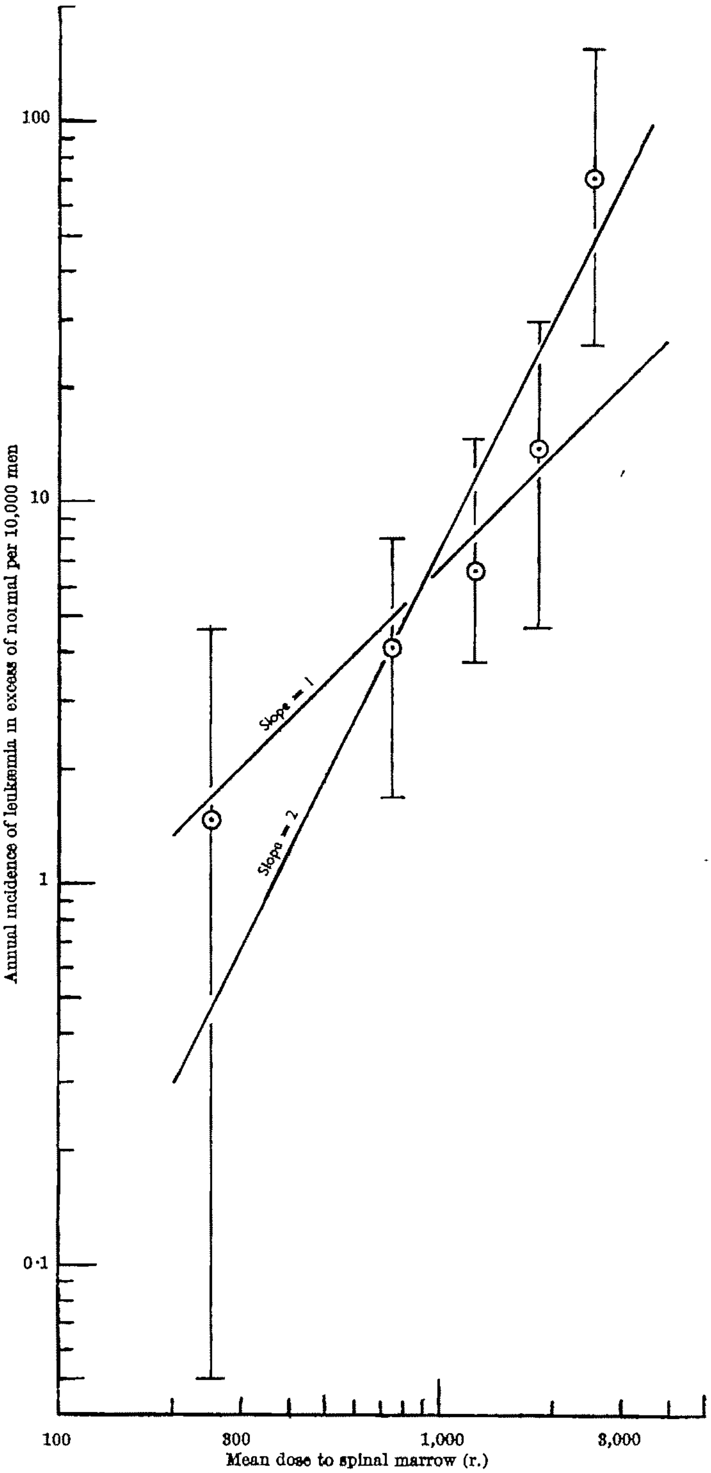

Fig. 1. Annual incidence of leukæmia in excess of normal per 10,000 men, versus mean dose (r.) to spinal marrow. Re-plotted from Court Brown and Doll (ref. 7) on log–log scales. Errors are taken from the original paper and represent 95 per cent confidence limits based simply upon the number of cases. The actual errors will be slightly larger

Further investigation will, however, be necessary to ascertain whether the classification 'bone tumours excluding jaw' refers predominantly to the same type of tumour as that induced by radium in bone. Nevertheless, for bone cancer, as for chronic myeloid

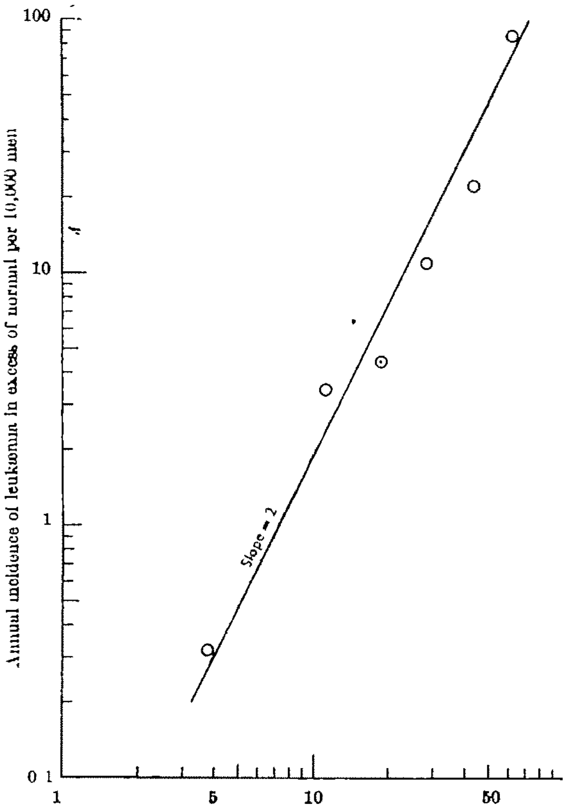

Fig. 2. Annual incidence of leukæmia in excess of normal per 10,000 men versus whole-body integral dose (Mgr.). Re-plotted from Court Brown and Doll (ref. 7) on log–log scales. The errors were not calculated, but they will be similar to, or slightly larger than, those in Fig. 1

leukæmia, it is possible that the natural incidence requires a two-mutation sequence and that radiogenic incidence requires a two-hit process.

Consider now the application of Fisher's theory to radiogenic cancer. Suppose the radiation dose is delivered over a long period (as with background or fall-out) and at a constant rate R rems per unit time.

Suppose m_1 = number of specific first mutations produced per rem, m_2 = number of specific second mutations per rem.

α = growth-rate constant for clones containing 'first-mutation cells'. (Fisher assumes for epithelial tissues that the number of cells in a clone increases in proportion to the square of the time, and we shall adopt the same growth law here.)

It can be shown that dN_2, the number of cells arising containing both specific mutations during a time interval t to $t + dt$ is given by:

$$dN_2 = \tfrac{1}{3}\,\alpha m_1 m_2\, R^2 t^3\, dt \qquad (1)$$

Since the total dose delivered is $D = Rt$,

$$dN_2/dt = \tfrac{1}{3}\,\alpha m_1 m_2\, D^2 t \qquad (2)$$

Thus, on this theory, if a two-mutation sequence is required for carcinogenesis, the rate of radiogenic tumour cell formation will be proportional not only to the square of the dose but also to the time over which it is delivered (provided t is large compared with the time of first-mutation cell division). In the same manner, widely spaced (that is, fractionated) doses should be extremely effective as compared with the same total dose delivered in a short interval.

We shall see later that, in spite of large uncertainties in the evidence, there is no indication that low dose-rates or fractionated doses possess this enhanced efficiency. Indeed, it is much more likely that low dose-rates are, in general, less effective than high.

Fisher's multiple mutation theory seems inapplicable, therefore, to radiation leukæmogenesis in humans, although where there is no cohort effect it offers an attractive interpretation of the dependence of the natural age-specific incidence-rate on age.

Summarizing, certain natural cancers may arise as a result of two or three successive somatic mutations, but radiogenic leukæmia and bone cancer may well originate from two discrete events which have certain properties in common with, but others different from, natural somatic mutations. One is, therefore, led to the conjecture that these discrete events in the radiation case could be chromosome or chromatid breaks. At the same time, it is necessary to assume that, after the first break, clone-growth does not occur, although in the eventual cancer cell the 'break' (gene deletion ?) must produce an effect similar to that of the natural mutation.

Chromosome-break Model

In this model it is assumed that a radiogenic cancer arises from a cell which has received two

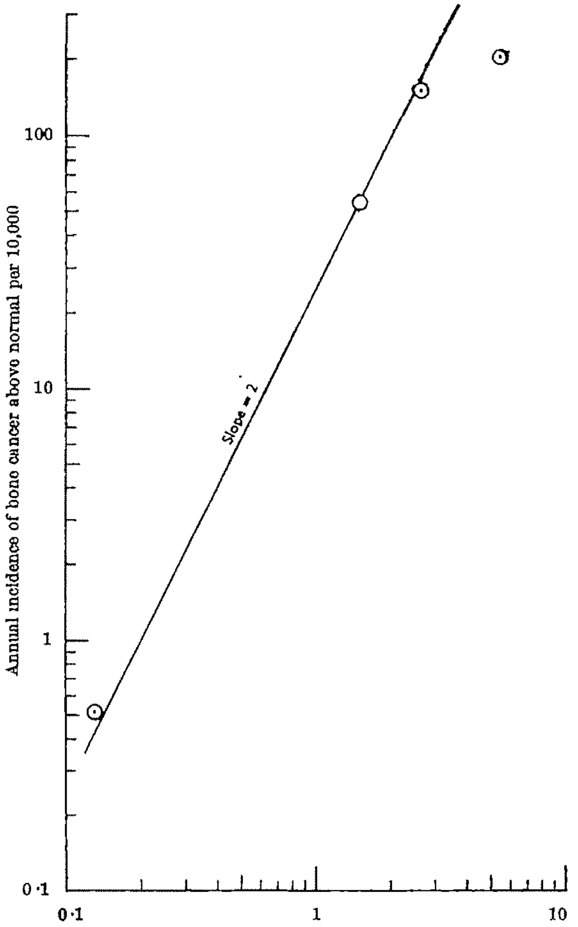

Fig. 3. Annual incidence of bone cancer above normal per 10,000 versus terminal radium burden (μgm. radium-226). Re-plotted on log–log scales from Marinelli (ref. 9). No error estimates were given in the original paper

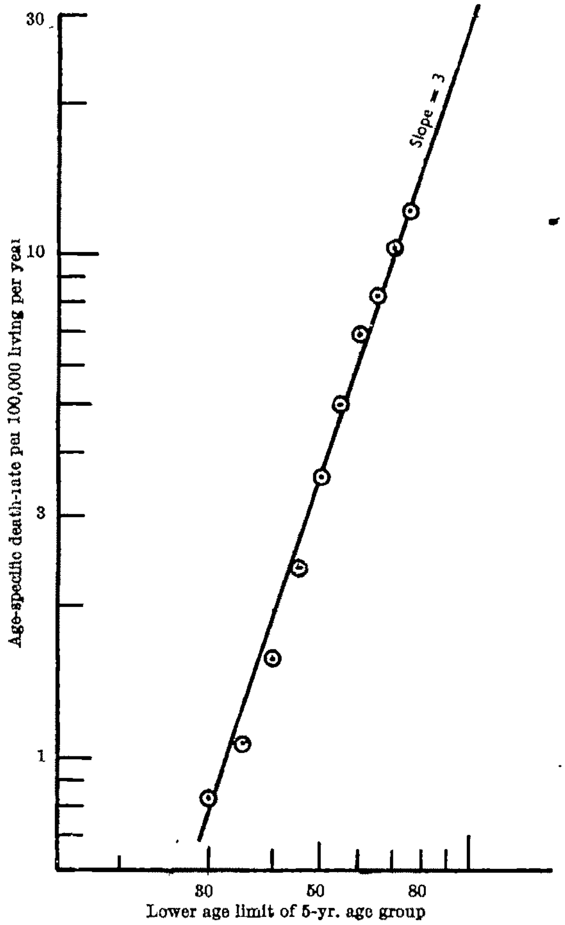

Fig. 4. Mean age-specific death-rate from cancer of bones (excluding jaw) for the 1866 and 1871 cohorts, versus age, on log–log scales. Plotted from Case's (ref. 13) analysis of statistics for England and Wales

specific chromosome breaks. The following detailed suppositions are made:

(*a*) L cells of a particular tissue type are capable of developing into cancer cells.

(*b*) They are irradiated at a constant dose-rate of R rems per unit time.

(*c*) c_1 is the average number of specific first chromosome breaks produced per cell per rem.

(*d*) Cells containing a 'c_1' break may be rendered carcinogenically ineffective through natural processes (such as repair or cell death), or through further radiation injury (such as another chromosome break) leading, perhaps, to cell death. The natural processes of elimination are characterized by a simple exponential function with decay constant λ (though the function could well be multiple exponential, Gaussian, or of some other form). p, the number of lethal radiation hits per 'c_1–type cell' per rem characterizes the radiation injury.

(*e*) c_2 is the average number of specific second chromosome breaks produced in a 'c_1–type' cell per rem.

(*f*) A long interval can elapse between the production of a potentially cancerous cell and the actual development of a cancer. If during this period, or a part of it, cancerous potentialities are carried by only one cell, the receipt of a further radiation 'hit' (producing, for example, another chromosome break) could be lethal to that cell. q is the number of such lethal hits per potentially cancerous cell per rem.

If the irradiation time is short so that a potentially cancerous cell cannot undergo division during the irradiation, it can be shown that the number, N_2, of potentially cancerous cells is given by:

$$N_2 = \frac{Lc_1c_2R^2}{\lambda + pR}\left\{\frac{1 - e^{-qRt}}{qR} + \frac{e^{-(\lambda + pR)t} - e^{-qRt}}{\lambda + R\,(p - q)}\right\} \quad (3)$$

(In solving the differential equations imposed by the assumptions, it is further assumed that at the commencement of the irradiation there are no c_1- or c_2-type breaks in any of the L cells.)

(*g*) If a potentially cancerous cell divides shortly after its production and cancerous potentialities are transmitted to both daughter cells, the chance of both cells being eliminated by subsequent radiation damage is very much reduced. If the 'vulnerable period' (τ) between the initial production of the potentially cancerous cell and its effective multiplication is short (such that $qR\tau \ll 1$), q-type lethal hits can be disregarded. For protracted irradiations under these conditions we obtain:

$$N_2 = \frac{Lc_1c_2R^2}{\lambda + pR}\left\{t - \frac{1 - e^{-(\lambda + pR)t}}{\lambda + pR}\right\} \quad (4)$$

The significance of equations (3) and (4) is best understood with reference to various limiting conditions. Consider first the dose–response relationship for small doses delivered at a high dose-rate, so that the following conditions obtain:

$$pR,\ qR \gg \lambda$$

$$\lambda t,\ pRt,\ qRt \text{ (that is, } pD \text{ and } qD) \ll 1$$

Equation (3) will be applicable and it reduces to:

$$N_2 = Lc_1c_2D^2/2 \quad (5)$$

If, however, a small dose is delivered at a low dose-rate over a very long period (as with background and fall-out radiations) such that $\lambda t \gg 1$, $\lambda \gg pR$,

$$pD,\ qD \ll 1$$

equation (4) reduces to:

$$N_2 = Lc_1c_2R^2t/\lambda = Lc_1c_2D^2/\lambda t \quad (6)$$

Under these conditions, cancer incidence will be less than that observed for the same dose delivered at high dose-rate in the ratio of $2/\lambda t$ ($\lambda t \gg 1$).

Consider now the effect of a large dose delivered at a high dose-rate in a short time,

$$\text{that is, } pD,\ qD \not\ll 1$$
$$pR \gg \lambda$$

Equation (3) is applicable, and it has been shown above for small doses at a high dose-rate that N_2 is proportional to D^2. As pD and qD increase with increasing dose, they will become an appreciable fraction of unity, and N_2 will fall below proportionality to D^2. In the limiting case where pD, $qD \gg 1$, N_2 reaches a plateau:

$$N_2 \rightarrow \frac{Lc_1c_2}{pq} \quad (7)$$

The remaining case to consider is the delivery of a large dose over a long period.

$$t \gg 1,\ \lambda t \gg 1,\ pD \gg 1$$

Equation (4) applies and it simplifies to:

$$N_2 = \frac{Lc_1c_2R^2t}{\lambda + pR} = \frac{Lc_1c_2D^2}{\lambda t + pD} \quad (8)$$

Before proceeding to a rough estimate of the constants in these equations, and in order to relate N_2 (total incidence) to observed incidence-rates, it is necessary to consider some of the possible biological processes involved in carcinogenesis.

Carcinogenesis : Possible Mechanisms

We have seen that on Fisher's multiple mutation theory the dependence of the natural age-specific incidence-rate on age for chronic myeloid leukæmia and for bone cancers can be understood if two successive somatic mutations are required within the same cell. It is also possible that radiogenic leukæmia and bone cancers result from two chromosome or chromatid breaks within the same cell : these breaks could produce gene deletion. It is tempting to conclude that the two mutations and the two breaks have a similar effect on the economy of the cell, and that in both cases the modified deoxyribonucleic acid will produce a modified ribonucleic acid synthesis. Modified ribonucleic acid will in turn affect protein synthesis in the cytoplasm.

On the basis of immunological studies, Green[14] contends that malignancy arises through the following stages : initially there is a modification of one or more cytoplasmic specific protein complexes (cell identity markers) ; these modified complexes then elicit an antibody response, to which the cell eventually 'adapts' by losing the modified marker. This loss of marker involves the failure of the normal growth-regulating mechanism and the 'adapted' cell becomes neoplastic. Other evidence points to a rather similar conclusion, that certain proteins essential to normal controlled division are either eliminated (corresponding to Green's antigenic loss) or structurally modified so as to be incapable of functioning normally[15].

Modified protein synthesis could result in irreversible injury of respiration which, Warburg[16] has claimed, is the essential first phase of cancer. The present hypothesis does not, however, predict such a specific form of injury. Thus, although there is no contradiction with Warburg's theory of cancer, the present hypothesis is, in its most important aspects, independent of it.

Osgood[17] argues that a basic dividing cell must generally give rise to one cell which proceeds to differentiate and another basic or primitive cell which is capable of maintaining this process through further division. This normal or 'arithmetic' type of division will occasionally (during growth or after injury) change to 'logarithmic' division in which two basic daughter cells emerge after cell division. Osgood postulates that logarithmic division is normally prevented by an inhibitor which, he suggests, is produced in the adjacent or nearly adjacent differentiated cells. Malignancy is said to result from a lowered concentration of inhibitor, and Osgood claims that a reduction in the life-span of differentiating cells could bring this about.

Although some such homœostatic control must exist, logarithmic growth could arise not only via Osgood's suggested mechanism but also through any appropriate interruption in the feed-back circuit and in particular through the malfunctioning of the 'inhibitor receptors' in the basic dividing cells. (These 'inhibitor receptors' may be equated with Green's cell identity markers.) If a unique two-hit process is involved in radiogenic leukæmia and bone cancer, it would seem that the initiating process is much more likely to involve a modification of the 'inhibitor receptors' than injury to one, or at the most two, differentiated cells. Osgood's explanation in terms of a reduced inhibitor concentration could, of course, apply in certain other situations where, for example, hormonal factors are important.

The biological mechanism of 'two-hit' radiation carcinogenesis favoured here is therefore as follows:

When a viable cell containing a c_1-type chromosome modification receives a c_2-type chromosome (chromatid) break, the resulting cell is potentially carcinogenic. Subsequent cell division will dilute the normal ribonucleic acid and the normal cell identity markers initially present until eventually a cell emerges in which normal markers have been entirely replaced by modified markers. At this stage (or possibly before) the antibody response postulated by Green will be elicited. Adaptation, involving the elimination of all markers, produces the carcinogenic cell.

The number of cell divisions required to attain the necessary dilution of cell markers will be affected by chance. Hence, an irradiation of a group of similar organisms should be followed by a minimum latent period, at the end of which the tumour incidence-rate should show an approximately normal or Poisson distribution in time.

The Hiroshima statistics[18] suggest that this may be true in the case of radiogenic human leukæmia, with peak incidence-rate occurring about seven years after irradiation (Fig. 5). On this hypothesis, the minimum latent period should also be independent of dose, except that heavy doses, by extensive cell destruction, could accelerate cell division among potentially malignant cells and produce a shortening of the minimum latent period. This relative independence of latent period on dose is also supported by the Hiroshima leukæmia statistics.

If the dose-range is low enough so that incidence (N_2) can be expected to be proportional to D^2 (that is, pD, $qD \ll 1$), then beyond the minimum latent period, cumulative tumour incidence and incidence-rate at a given time after the commencement of irradiation should both be proportional to the square of the dose.

Brues's experiments[19], in which the daily probability of bone tumour appearance in mice was observed as a function of strontium-89 dose, support this contention, although previously they have been differently interpreted. Where there is a time overlap between incidence-rate curves at different doses, the probability of incidence at a given time is, within experimental error, approximately proportional to (dose)^2. This is true for both continuous and single injection experiments. The apparent lengthening of the latent period at the lower doses is therefore due, in part at least, to inadequate statistics, for when doses differ by a factor of 10, incidence will differ by a factor of 100 ; consequently, significant increases above the normal or spontaneous incidence emerge much sooner at high doses.

Finkel and Biskis[20], also on the evidence of experiments with mice, conclude that the latent period for the induction of osteogenic sarcomas is independent both of isotope and of dose. If this deduction is true, then according to the present hypothesis two consequences should follow. First, in their Fig. 4, where the slope of the changing tumour-rate is plotted against injected dose (on log–log scales), the curves should have a slope of 2 over the lower dose-ranges. (This follows if N_2 is proportional to the square of the dose and if tumour-rate is distributed in time according to an error function the width of which is independent of dose.) For strontium-90 the slope is slightly less than 2, but for calcium-45 it is slightly more than 2; the data for radium-226 are too few to allow the (steep) slope at low dose to be determined, but at high dose the slope becomes less than unity, implying, as would be expected, that p and q for α-particles are somewhat greater than for β-particles. Secondly, the cumulative incidence of osteogenic sarcomas before death intervenes should at a given time (and at low dose) be proportional to the square of the dose. Judging from Finkel and Biskis's Fig. 1, this relationship, in general, does not hold, although the discrepancy could be explained if, contrary to their deductions, the latent period at high dose is, in fact, reduced.

In connexion with these experiments on mice, it should be mentioned that Marinelli[9] has estimated that at low doses the relative biological effectiveness of α-particles is 4. It is interesting to inquire what combination of radiation damage to the chromosome material could yield this relative biological effectiveness. Some important data concerning the frequency of chromosome and chromatid breaks are assembled in Lea's[31] Table 67, and although they refer to *Tradescantia* microspores they will be used here as a guide. If a two-hit mechanism in the organism consisted of one chromatid break which joined and one which remained unjoinable, the relative biological effectiveness of α-particles (relative to X-rays) can be deduced from Lea's table to be approximately 4.

If radiogenic bone tumours in mice were produced by somatic mutations rather than through breaks in the chromosome material, we should expect the relative biological effectiveness of α-particles to be less than unity. The relative biological effectiveness evidence for radiogenic bone tumours in mice is therefore consistent with the chromosome (chromatid) break hypothesis and at variance with the multiple-mutation theory.

It is not intended to suggest that the present 'two-hit' or 'double-break' hypothesis is applicable to all cases of radiation carcinogenesis; thus in mice, radiogenic thymic leukæmia and ovarian neoplasia may very well be produced by a 'threshold' or generalized-damage mechanism. I hope to discuss the 'threshold' concept on another occasion.

Application of Chromosome-break Hypothesis to Human Leukæmogenesis

The numerical results derived in this section must be treated with the utmost reserve since uncertain data are substituted in an extremely tentative hypothesis. It may be said, however, that the same is true of all the previous estimates of radiogenic leukæmia incidence.

The Hiroshima and Nagasaki leukæmia statistics would be of immense theoretical importance if it were possible to estimate accurately the dose to the survivors. In the absence of detailed information, an attempt has been made to obtain very approximate averages for the Hiroshima survivors using the following procedure. Air doses for γ-rays and fast neutrons were taken from curves prepared by the Health Physics Division of Oak Ridge National Laboratory[32]. The mean attenuation of γ-rays is assumed to be 0·76 in buildings and 0·74 in tissue overlying bone marrow. The mean attenuation of fast neutrons is assumed to be 0·48 in buildings and 0·26 in tissue overlying bone marrow; the relative biological effectiveness for fast neutrons is taken to be 2. Fatalities were very high in the inner zone (0–1,000 m. from the hypocentre) and most of the leukæmia victims were in the region 850–1,000 m.[3]. Here, the average bone marrow dose (assuming a uniform geographical distribution of the population) is found by integration to be 600 rems. Similarly for the zone 1,000–1,500 m., the average is 170 rems. For the zone 1,500–2,000 m., where incidence is expected to be proportional to (dose)^2, the mean square dose has been calculated and found to be nearly 700 (rems)^2; the root mean square dose is, therefore, 26 rems. The mean or root mean square doses are plotted in Fig. 6 versus the mean leukæmia incidence-rate[18] for the period 1950–57 on log–log scales. The points are consistent with the type of relationship predicted by equation (3). That is, at low doses, incidence is proportional to (dose)^2, but at high dose the curve bends over and attains a plateau.

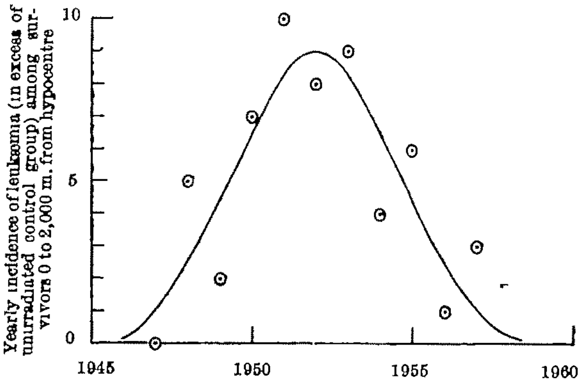

Fig. 5. Yearly incidence of leukæmia in excess of unirradiated control group, among the survivors at Hiroshima who were 0–2,000 m. from the hypocentre, versus year. Plotted from Wald (ref. 18).

If the incidence of radiogenic leukæmia for the lowest dose group is distributed in time according to the smooth curve of Fig. 5, and if no other peaks occur in future years, the total incidence of radiogenic leukæmia for this group is calculated to be 1·5 per 10,000.

From equation (5):

$$Lc_1c_2 = \frac{2N_2}{D^2} = \frac{3\cdot0 \times 10^{-4}}{694}$$
$$= 4\cdot3 \times 10^{-7}$$

Brues[1], on the assumption that leukæmia arises from proliferating myeloid cells, has suggested that L is of the order of 10^{11} cells.

$$\text{Hence } c_1c_2 \sim 4 \times 10^{-18}$$

Assume that c_1 and c_2 refer to two chromatid breaks, one of which is restituted and the other remains broken. If a fraction 0·09 of primary breaks are unjoinable[31], then if c_1 and c_2 are now referred to primary breaks,

$$(c_1c_2)_{\text{primary breaks}} \sim \frac{4 \times 10^{-18}}{0\cdot09 \times 0\cdot91}$$
$$\sim 5 \times 10^{-17}$$

Since c_1 and c_2 are likely to be of the same order of magnitude:

$$c_1 \sim c_2 \sim 7 \times 10^{-9} \text{ specific primary breaks/rem}$$

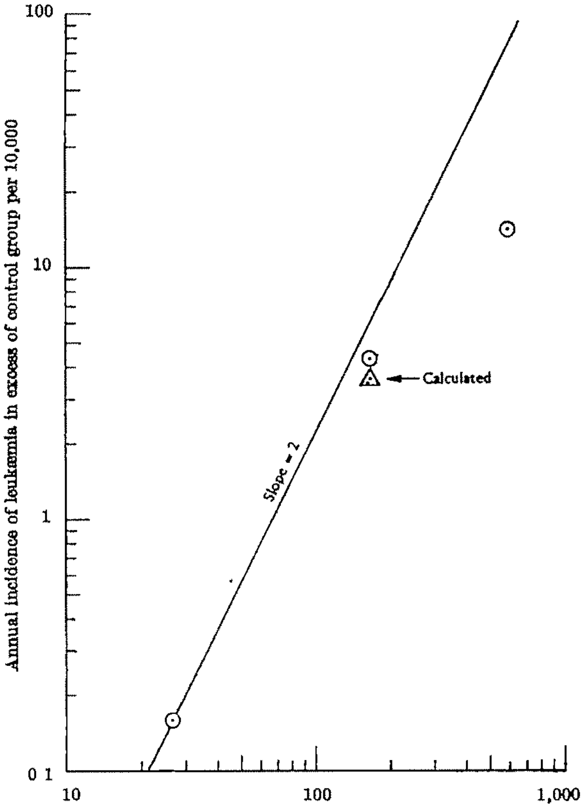

Fig 6. Mean annual incidence of leukæmia at Hiroshima above unirradiated control group per 10,000 for 1950–57, versus mean, or root-mean-square, dose to bone marrow (log–log scales). The leukæmia statistics (circles) are taken from Wald (ref. 18) and the doses are calculated from Ritchie and Hurst (ref. 22) (see text). The point enclosed in a triangle is calculated from the present hypothesis (see text)

On the basis of Lea's target theory[21], this figure is found to correspond to a target molecular weight for each specific primary break of the order of 10^4. This reasonable value contrasts with the absurdly small target volumes entailed by the linear theory of carcinogenesis and commented on by Brues[1].

It is also apparent that if a tumour required for its initiation the co-existence of three specific chromosome (chromatid) breaks in one cell, then for any reasonable values of L, c_1, c_2 and c_3 the chance of a radiogenic tumour arising from doses of the order of 1,000 rems must be exceedingly small. Thus, if and when radiogenic tumours arise from specific modifications to the chromosome material of a single cell, only those tumours are likely to be observed which require two such modifications. (It is possible, however, to account for the occurrence of natural tumours requiring three successive mutations on the hypothesis of clone-growth of mutated cells.)

For the point at highest dose in Fig. 6 it can be shown that equation (7) is a good approximation, hence:

$$pq = \frac{Lc_1c_2}{N_2}$$

From Wald's[18] statistics and the smooth curve of Fig 5, $N_2 = 1{\cdot}39 \times 10^{-2}$ leukæmias per person.

Therefore,

$$pq = \frac{4{\cdot}3 \times 10^{-7}}{1{\cdot}39 \times 10^{-2}} = 3{\cdot}1 \times 10^{-5}$$

If, as is probable, p and q are of the same order of magnitude, $p \sim q \sim 6 \times 10^{-3}$ lethal hits per cell per rem.

This figure may be compared with the values $7{\cdot}2 \times 10^{-3}$ chromatid breaks per cell per r., and 6×10^{-4} chromosome breaks per cell per r., quoted by Lea[21] for the production of aberrations in *Tradescantia* microspores by high-energy X-rays.

The expected incidence-rate for the mid-point in Fig. 6 can now be calculated from the constants derived above, if it is assumed that p is not identically equal to q. (Since R here is very large, it is highly probable that in equation (3), $R\ (p - q) \gg \lambda$. To achieve this it must be assumed, $p \not\equiv q$.) Fortunately, the value of N_2 is not critically dependent on p and q, and putting $p = 6 \times 10^{-3}$, $q = 5 \times 10^{-3}$, we obtain $3{\cdot}6 \times 10^{-4}$ leukæmias per person per year (for the years 1950–57), which is in satisfactory agreement with the observed value of $4{\cdot}3 \times 10^{-4}$.

Finally, the statistics for leukæmia incidence among American radiologists enable a crude estimate of λ to be made. It must, however, be emphasized that differences (for example, in age and sex distributions) between this group and the Hiroshima survivors make the value of such a derivation somewhat dubious. Over the period 1938–52 the average death-rate among American radiologists from radiogenic leukæmia was approximately 4·9 per 10,000 per annum[3]. Deaths within the age-range 35–74 years were recorded and the average expectation of life was 62 years. Assuming equilibrium conditions, then for radiologists older than 35 years, the total expectation of radiogenic leukæmia would have been approximately $(62-35) \times 4{\cdot}9 \times 10^{-4}$, that is, $1{\cdot}32 \times 10^{-2}$. The mean total exposure time to radiation was 24·4 years[3]. Braestrup estimates that the typical exposure dose-rate to radiologists working under poor protection conditions was about 100 r. per annum, corresponding to a dose-rate to the bone marrow of 66 rems per annum[23]. With the subsequent improvement in protection, the marrow dose-rate has now fallen to less than 1 rem per annum. Assuming that equation (8) is applicable, we may take R to be (very roughly) 50 rems per annum. Solving for λ, we obtain the value $1{\cdot}_{75}$ yr.$^{-1}$, but since it depends on almost the square of the (uncertain) dose-rate, the error could, perhaps, amount to one order of magnitude. (This assumes that we are dealing with a simple exponential function—and this assumption might well be false.)

Predicted Incidence of Radiogenic Leukæmia at Low Dose-rates

It is interesting to apply this hypothesis to the incidence of radiogenic leukæmia to be expected from background and fall-out radiations.

Background radiation. The following assumptions are made:

$$R = 0{\cdot}1 \text{ rem/yr.} \quad t = 70 \text{ yr.}$$

$$\frac{Lc_1c_2}{\lambda} = \frac{4{\cdot}3 \times 10^{-7}}{1{\cdot}75} = 2{\cdot}5 \times 10^{-7}$$

(In the absence of better information this is taken to be constant throughout life, although obviously L, at least, will vary considerably.)

Applying equation (5), $N_2 \sim 1{\cdot}7 \times 10^{-7}$.

Thus, neglecting problems of latent period, in a population of 50 million people of average life-expectancy 70 years, background radiation may

Table 1

Radiation	Dose-rate assumed (rems/yr.)	Duration of exposure (yr.)	Incidence-rate of radiogenic leukæmia		
			Linear theory (after Lewis, ref. 3)	Present 'two-hit' theory. Possible range	
Natural background	0·1	70	25,000	1 to 100	per 50 million persons in 70 years (as compared with nearly 200,000 'natural' cases)
Fall-out	0·01	70	2,500	0·2 to 20	
Maximum permissible occupational exposure	5	40	32	0·1 to 10	in a population of 4,000 over a life-time of occupational exposure (as compared with 15 'natural' cases)

The effect of low dose-rate ionizing radiations on radiogenic leukæmia incidence in humans. A comparison between predictions made (*a*) on the basis of the linear theory (after Lewis, ref. 3) and (*b*) by the 'two-hit' hypothesis presented here. For reference, the 'natural' total incidence based on present statistics for England and Wales is shown in the last column

induce of the order of ten cases of radiogenic leukæmia over a 70-yr. period.

This is exceedingly small compared with the 'natural' incidence-rate in Great Britain of about fifty cases per million persons per annum. It is also very small compared with the best predictions made with the linear theory. Thus, Lewis[3] estimates for the 'average' individual in a population that the probability of radiation-induced leukæmia is 2×10^{-6} per rem per annum. Hence, in a population of 50 millions, the total number of radiogenic leukæmias appearing over a 70-yr. period for a dose-rate to each individual of 0·1 rem per annum would be:

$$\simeq 5 \times 10^7 \times 2 \times 10^{-6} \times 0{\cdot}1 \times \frac{70^2}{2}$$

= 25,000 (as compared with a possible range of 1–100 predicted above).

Fall-out radiation. A dose-rate to the individual from internal and external sources of 0·01 rem/yr. will be assumed (this is probably in excess of contemporary dose-rates from fall-out). If this is added to the normal background, and if the influence of other sources of radiation is ignored, the additional incidence, ΔN_2 of radiogenic leukæmia is given by:

$$\Delta N_2 = \frac{Lc_1c_2t}{\lambda}(0{\cdot}11^2 - 0{\cdot}10^2)$$
$$\sim 3{\cdot}6 \times 10^{-8}$$

(According to this hypothesis, ΔN_2 depends upon the normal background dose-rate.) Thus, in a population of 50 million people of average life-expectancy 70 years, an increase of the radiation background from 0·1 to 0·11 rem/yr. should produce of the order of two additional cases of radiogenic leukæmia over a 70-yr. period. Lewis's relationship predicts 2,500 cases under these conditions (Being based on a linear theory, this figure is unaffected by the level of natural background radiation.)

Maximum Permissible Occupational Exposure Dose

In the United Kingdom, the maximum permissible life-time dose to the blood-forming organs of radiation workers is 200 rems. If this dose is spread uniformly over forty working years ($R = 5$ rems/yr.) we find from equation (3) that $N_2 \sim 2{\cdot}5 \times 10^{-4}$ leukæmias per person. Hence, in a population of 4,000, exposed for an occupational life-time at the maximum permissible dose-rate, we should, according to this hypothesis, expect of the order of one radiogenic leukæmia to be induced. On the basis of Lewis's[3] linear relationship, the corresponding figure would be thirty-two cases; at this higher dose-rate the disparity between the two theories is much narrower.

Osteogenic Sarcoma

The available data relating to human osteogenic sarcoma are too scanty to enable numerical predictions to be made for low dose-rate conditions with the present hypothesis.

Before concluding, it should be mentioned that alternative two-break hypotheses can be entertained. We can, for example, conceive a model in which $\lambda = 0$ and c_1 and c_2 are functions of dose-rate (R). Starting from the same statistics of human radiogenic leukæmia, the predictions from such a model would probably not differ greatly from the predictions made here. However, there is no obvious way at present of establishing a relation between c_1, c_2 and dose-rate.

Finally, any plausible alternative to the linear theory is likely to predict a very much lower incidence of radiogenic tumours at low doses and low dose-rates. It is difficult to imagine a realistic hypothesis which would lead to predictions intermediate between linear (one-hit) and quadratic (two-hit) theories, while any 'threshold' theory would be likely to involve an altogether negligible incidence at dose-rates comparable to background.

[1] Brues, A. M., *Science*, **128**, 693 (1958).

[2] Kimball, A. W., *J. Nat. Cancer Inst.*, **21**, 383 (1958).

[3] Lewis, E. B., *Science*, **125**, 965 (1957).

[4] Mewissen, D. J., *Nature*, **183**, 401 (1959).

[5] Finkel, M. P., *Science*, **128**, 637 (1958).

[6] Newcombe, H. B., *Science*, **126**, 549 (1957).

[7] Court Brown, W. M., and Doll, R., Medical Research Council, Spec. Rep. No. 295 (1957).

[8] Report of the United Nations Scientific Committee on the Effects of Atomic Radiation; General Assembly, Official Records: Thirteenth Session Supplement No. 17 (*A*/3838), New York (1958).

[9] Marinelli, L. D., *Amer. J. Roentgenol.*, **80**, 729 (1958); and private communication.

[10] Muller, H. J., *Sci. in Prog.*, **7**, 130 (1951). Nordling, C. O., *Nord. Med.*, **47**, 817 (1952). Armitage, P., and Doll, R., *Brit. J. Cancer*, **8**, 1 (1954); **11**, 161 (1957). Failla, G., *Radiology*, **69**, 23 (1957).

[11] Fisher, J. C., *Nature*, **181**, 651 (1958).

[12] Court Brown, W. M., and Doll, R., *Brit. Med. J.*, **i**, 1063 (1959).

[13] Case, R. A. M., *Brit. J. Prev. and Soc. Med.*, **10**, 172 (1956).

[14] Green, H. N., *Brit. Med. Bull.*, **14**, 101 (1958).

[15] Miller, J. A., and Miller, E. C., "Adv. Cancer Res.", **1**, 339 (1953). Haddow, A., *Brit. Med. Bull.*, **14**, 79 (1958).

[16] Warburg, O., *Science*, **123**, 309 (1956).

[17] Osgood, E. E., *J. Nat. Cancer Inst.*, **18**, 155 (1957).

[18] Wald, N., *Science*, **127**, 699 (1958).

[19] Brues, A. M., *J. Clin. Invest.*, **28**, 1286 (1949).

[20] Finkel, M. P., and Biskis, B. O., *Acta Union Internationale contre le Cancer*, **15**, 99 (1959).

[21] Lea, D. E., "Actions of Radiations on Living Cells" (Cambridge University Press, 1946 and 1955).

[22] Ritchie, R. H., and Hurst, G. S., *Health Phys.*, **1**, 390 (1959).

[23] Braestrup, C. B., *Amer. J. Roentgenol.*, **78**, 988 (1957); and private communication.

OBITUARIES

Dr. R. A. Frazer, F.R.S.

The news of the death of Dr. R. A. Frazer at his home in Ockham on December 10 was received with much sadness by his former colleagues at the National Physical Laboratory, and particularly by those who had had the privilege of working with him or under his leadership. His unselfishness, charm, modesty and humour made him a most likeable person and he will be sadly missed by many friends. His scientific ability and distinguished bearing inspired respect in all who met him, and his kindliness won the affection of all those who had the good fortune to know him well.

Dr. Frazer had a brilliant career as a mathematician at Cambridge and was a Rayleigh Prizeman. He joined the staff of the National Physical Laboratory in 1914, his first post being that of assistant secretary in administration. After about a year he was transferred to a group engaged in aerodynamic research in the Engineering Division. This group of pioneers in aeronautics became the nucleus around which the Aerodynamics Division was formed a few years later.

During the first ten years or so of Frazer's career as a research worker, his mathematical talent was put to good use in the study of problems of viscous flow, but from time to time he took part in other investigations. Some of the work he did in connexion with full-scale tests on the airship *R*.38 resulted in his winning the *R*.38 Memorial Prize awarded by the Royal Aeronautical Society.

It was characteristic of him to do everything he attempted with great thoroughness, and he was never satisfied until he had unravelled all aspects of any problem, however complicated, on which he happened to be engaged. He had remarkable power for concentration and when deeply immersed in a problem would lose all sense of time.

It was not surprising that, when aircraft flutter became a serious problem around about 1924, Frazer was asked to investigate what was then a mysterious phenomenon. By wind tunnel tests on simple aircraft models of reduced elasticity, and by analysis, he and his co-workers were soon able to obtain a physical understanding of flutter and to discover methods for its prevention. With W. J. Duncan as co-author he published in 1928 the first flutter monograph, and this is now often referred to as the 'flutter bible'. In this book the phenomenon is clearly explained by reference to simple systems, and the basic theory for the prediction of critical speeds for flutter is fully developed. His study of flutter theory led him to take an interest in matrix algebra for the purpose of analysing complicated systems involving many degrees of freedom. The outcome of this work was a book on "Elementary Matrices", written with Duncan and Collar as co-authors. This is now regarded as a classic on the subject, and it has had a profound world-wide influence on the development of methods of flutter and vibration analysis and on the general theory of aero-elasticity.

In recognition of his outstanding work he was elected a Fellow of the Royal Society in 1946. He was also a Fellow of the Royal Aeronautical Society and of the Institute of the Aeronautical Sciences, and was a member of the Oscillation Sub-Committee of the Aeronautical Research Council. Formerly he was also a member of the Mechanics Committee.

Immediately after the Second World War, the National Physical Laboratory was asked to investigate the possibility of wind-excited oscillations of suspension bridges, in connexion with the proposal to erect suspension bridges across the Severn and the Forth. It was feared that, like the Tacoma bridge, these bridges would fail unless they were properly designed to avoid such instability. Again it was Dr. Frazer who took responsibility for the investigation. With the assistance of C. Scruton and other members of the Division, he designed and made a 50-ft. long reduced elasticity model for testing in a specially erected wind tunnel at Thurleigh. In due course they were able to reproduce on the model the kind of oscillation that destroyed the Tacoma bridge and show how it could be avoided by suitable design. As a result of their investigations they were able to propose a design for the Severn and Forth Suspension Bridges which was not prone to instability in the strongest winds. A bridge to this design is now being built over the Firth of Forth.

Outside his work, Dr. Frazer had many interests. In his younger days he was a keen mountaineer and enjoyed nothing better than climbing in the Alps, the Lake District and North Wales. He was also a member of Oxford University Expeditions to Spitsbergen in 1921, 1923 and 1924. In later years his main interest was gardening. When he retired in 1954 after forty years service, everyone hoped that he would have good health to enjoy a long and happy retirement with his family, but this was not to be.

Dr. Frazer's name will live long in aeronautical history, and he will always be remembered with esteem and affection by many friends who feel the deepest sympathy with his wife and family in their bereavement W. P Jones

Prof. Anrezej Soltan

The death has been announced of Dr. Anrezej Soltan, director of the Polish Institute of Nuclear Research since 1955. Born in 1897, Dr. Soltan was a member of the Scientific Council, Joint Institute for Nuclear Research, Dubna, U.S.S.R., a member of the Polish Academy of Sciences, and he held the position of chairman of the Central Administration of the Polish Physical Society during 1952–54.

Soltan was educated at the University of Warsaw and carried out scientific work in France during 1927–28 and in the United States during 1932–33. His publications cover the fields of spectroscopy, physical chemistry of hydrogen and mercury vapour, the interaction of fast neutrons with atomic nuclei and the isomeric radioisotope of bromine.

After the War he became professor at Lodz Polytechnic (1945–47), returning to the University of Warsaw as professor of experimental physics (1947–48) and then as professor of atomic physics.

Dr. Soltan was a delegate at the Second International Conference on the Peaceful Uses of Atomic Energy at Geneva in 1958 and was a member of the State Council for the Peaceful Uses of Nuclear Energy.

NEWS and VIEWS

The Agricultural Research Council: Sir William K. Slater, K.B.E., F.R.S.

Sir William Slater will be retiring on June 30 from the secretaryship of the Agricultural Research Council, a post which he has filled with great distinction during the past eleven years. On graduating in chemistry at the University of Manchester, Sir William did research under Prof. Lapworth and for three years he was an assistant lecturer in the University. After two years in industry, he joined Prof. A. V. Hill at Manchester to assist in his work on muscle physiology. In 1923 he was awarded a Beit Memorial research fellowship and worked in the Department of Biochemistry of University College, London, under Sir Jack Drummond, where he developed a pioneer line of research on the biochemistry and physiology of insects, particularly anaerobiosis. In 1928 he went to Dartington Hall as scientific adviser to the trustees. Afterwards, he took charge of the agricultural and rural development on the estate and established a laboratory for applied research. In 1943, Sir William joined the staff of the Ministry of Agriculture, Fisheries and Food as secretary of the Agricultural Improvement Council for England and Wales, which had just been set up. He became responsible for the development of that Council's work, and had much to do with fostering the close relationship which exists between it and the Agricultural Research Council. Later, he became in addition senior advisory officer, responsible for setting up the specialist scientific section of the National Agricultural Advisory Service. This broad experience qualified Sir William well for the post of secretary of the Agricultural Research Council to which he was appointed in 1949. During his term of office he has brought to a successful conclusion a ten-year post-war programme for the development of agricultural research approved by the Cabinet in 1945. In 1947–48 the amount spent by the State on agricultural research was of the order of one million pounds; to-day it amounts to about six million pounds. New institutes have been created and existing ones have been expanded. Links with universities have been strengthened by the establishment of a number of units in university departments. The Council has indeed been fortunate in having the services of Sir William Slater over this period, and his wise guidance and never-failing patience will be missed.

Prof. E. G. Cox, F.R.S.

Prof. Ernest Gordon Cox, who will be succeeding Sir William Slater as secretary of the Agricultural Research Council on July 1, 1960, comes originally from Somerset. He was born at Bath in 1906 and educated at the City of Bath Boys' School and the University of Bristol, from which he graduated with first-class honours in physics in 1927. From Bristol he went as research assistant to Sir William Bragg at the Royal Institution during 1927–29 and there began the series of researches which transformed his formal field of interest from physics to chemistry. During 1929–45 he held appointments in the Department of Chemistry of the University of Birmingham, and in 1945 was appointed professor of inorganic and physical chemistry at the University of Leeds; his title was changed in 1953 to professor of inorganic and structural chemistry in a perhaps hopeless attempt to define more precisely the many-sided implications of his researches. He was one of the first to apply X-ray methods to solve problems in the stereochemistry of metal co-ordination compounds. But he has also made many contributions in organic chemistry, from his early work on α-chitosamine hydrochloride and hydrobromide, which established, by direct X-ray analysis, the existence and form of the pyranose ring in sugars, to his latest, very accurate, studies, with D. W. J. Cruikshank and others, of the electron distribution in benzene. In recent years his School of Chemistry has made Leeds an international centre to be visited by all interested in exact X-ray analysis. At the same time he has had much to do with problems of organization and administration, first in war research and later, in Leeds City and University; for the last three years, as chairman of the X-ray Analysis Group, he has been concerned with the practical applications of X-ray methods in many different fields. His variety of experience and knowledge, his own personal interest in agricultural problems (he is a serious amateur botanist and ornithologist) and his generous spirit should be of the greatest value in guiding agricultural research.

Building Science at Sheffield: Prof. J. K. Page

Mr. J. K. Page, at present lecturer in environmental physics in the Department of Building Science of the University of Liverpool, has been appointed to the newly created chair of building science in the University of Sheffield. Mr. Page was educated at Haileybury College and the University of Cambridge, where he gained first-class honours in Part I of the Natural Sciences Tripos. After graduation, Mr. Page held a temporary appointment with the Council of Industrial Design, where he assisted with plans for the Festival of Britain. After teaching at Westminster School for two years he took an appointment as senior scientific officer in the Colonial Liaison Section at the Building Research Station. During this appointment Mr. Page travelled extensively in West Africa, America and the West Indies. After a period as chief research officer of the Nuffield Foundation Division of Architectural Studies, Mr. Page was appointed to his present post in the University of Liverpool in 1957.

Royal Astronomical Society Awards

Awards of medals have been made to the following by the President and Council of the Royal Astronomical Society: *Gold Medal* to Prof. V. A. Ambartsumian, president of the Academy of Sciences and director of the Byurakan Observatory, Armenia, for his numerous important contributions to both theoretical and observational astronomy; *Eddington Medal* to Dr. R. d'Escourt Atkinson, deputy chief scientific officer at the Royal Greenwich Observatory, for his pioneering papers on atomic synthesis and stellar energy; *Jackson Gwilt Medal and Gift* to Mr. F. Maine Bateson, of Cook Islands, and Mr. A. F. A. Lofley Jones, of Timaru, New Zealand, for their work on variable stars in the southern hemisphere.

British Association: Installation of President

The installation on January 8 of Sir George Thomson as president of the British Association for 1960 (see *Nature*, 184, 151; 1959) marked the close of a year of increasing activity and encouraging and significant developments. The lecture service, in its infancy in January 1959, is now well under way. Some 500 scientists have agreed to give lectures, and their audiences run into five figures—around 22,000 adults from trade unions, rotary clubs, women's institutes and chambers of commerce, and 32,000 children from all types of schools. The three pioneer area committees of 1958 have been joined by twelve more; the last of these, in York, is a direct follow-up of the recent annual meeting there. The decision to make a number of fundamental changes in the quarterly journal of the Association was also taken during the past year. From May 1960 *The Advancement of Science*, in a completely revised format, will be published six times a year, instead of four. Though still containing papers delivered at the annual meeting, the journal will include much new material, and will be of an essentially non-specialist character, aimed to appeal to scientists working in other fields, students and sixth forms in the schools, and laymen who are anxious to understand something of the significance of scientific research and development. The wide potential circulation of such a journal should make it a powerful weapon in the Association's campaign.

Nuclear Energy Conferences and Symposia in 1960

The International Atomic Energy Agency, Vienna, has issued a provisional schedule of meetings to be organized in 1960. One large conference will deal with Radioisotopes in Physical Sciences and Industry and will probably be held at the end of August. The last major isotope conference, apart from the sessions devoted to this subject at the Geneva Atoms for Peace Conference in September 1958, was organized in Paris in September 1957 by Unesco, which will co-operate in the 1960 isotope conference. Another scientific conference will deal with problems of medium and small-size reactors and is planned for the beginning of September in Vienna with the expected participation of the United Nations. The tentative list of symposia to which invitations will be extended to all the Agency's Member States is as follows: (1) Fuel Element Fabrication with Special Emphasis on Cladding Materials (May 3–6, in Vienna); (2) Radiation Dosimetry in Physical Sciences (June 7–13, in Vienna); (3) Effects of Ionizing Radiation on Seeds and its Significance on Crop Improvement (probably in Karlsruhe, August 8–12, in co-operation with the Food and Agriculture Organization); (4) Inelastic Scattering of Neutrons in Solids and Liquids (October 11–14, in Vienna); (5) Neutron Pile Research (October 17–21, in Vienna); (6) Chemical Effects of Nuclear Transformation (October 24–27); (7) Nuclear Ship Propulsion with Special Reference to Safety (mid-November; the arrangements will be worked out in consultation with the International Maritime Consultative Organization); (8) Experimental and Test Reactors (November 28–December 2, in Vienna); (9) Pest Control by Radiation (probably in India in December, in co-operation with the Food and Agriculture Organization); (10) Use of Radioisotopes in the Study of Endemic and Tropical Diseases (in the Far East at the end of November or beginning of December, in co-operation with the World Health Organization); (11) Education and Nuclear Energy (probably in Argentina at the end of the year or beginning of 1961, in co-operation with Unesco).

Scientific Man-power in Britain

In an article, "The Statistics of Scientific Manpower", in the January 7 issue of *Education*, Sir Solly Zuckerman points out that the figures since published by the Scientific Man-power Committee, which were not available to Prof. J. Jewkes when he presented his analysis of the marked shortage of scientists before Section *F* (Economics) of the British Association at York last September, indicate that the conclusion drawn by Prof. Jewkes is wide of the mark. Graduates in pure science from British universities during 1956–59 numbered only 15,500, and not 18,000 as he stated, and even taking into account about 800 scientists, not considered by Prof. Jewkes, who were admitted to membership of the Royal Institute of Chemistry or the Institute of Physics on the strength of qualifications other than a university degree, when allowance is made for those who returned to their homes overseas or took up employment abroad (about 500) and for the 5,000 needed to replace losses, we are left with about 10,800 to satisfy any net increase in demand expected between 1956 and 1959 (11,700 in the 1956 estimate).

The Scientific Man-power Committee's recent report also shows that there were still about 4,000 vacancies at the beginning of 1959. The new estimates made by employers themselves of the number of scientists they will require by 1962 suggest that the general market shortage of scientists is still likely to be about 2,000 in 1962. Commenting further on Prof. Jewkes's 'working rule' that employers know best about their future requirements, Prof. Zuckerman also points out that the demand between 1956 and 1959 rose much more sharply than employers anticipated, and that in practically no branch of industry did action follow intention. We have still far to go before we need be unduly concerned about the exhortatory character of Prof. Jewkes's concept of 'unmet need'; but the Scientific Man-power Committee would certainly agree wholeheartedly with him that the criteria now available for estimating how many scientists and technologists will be needed in the long-term are very crude and urgently need to be refined.

Computation and Automatic Control

At the annual general meeting of the British Group for Computation and Automatic Control of the British Conference on Automation and Computation, held on December 17, 1959, the Executive Committee was reconstituted for 1960 as follows: *chairman*, Mr. J. F. Coales; *vice-chairmen*, Mr. R. Hindle and Mr. H. W. G. Gearing; *honorary treasurer*, Mr. S. M. Rix; *honorary secretary*, Mr. W. Bamford; *members*, Dr. A. D. Booth, Mr. N. Clarke, Mr. E. C. Clear Hill, Mr. W. C. F. Hessenberg, Mr. N. C. Pollock, and Prof. E. J. Richards. Groups *A* and *B* have undertaken the sponsoring of the British contribution to the First Congress of the International Federation of Automatic Control which is being held in Moscow during June 27–July 6. Further details can be obtained from the Honorary Secretary, Group *B*. Arrangements are being made also for a British Convention on this subject to be held in London in September, and further information can be obtained from the Honorary Secretary, Group *A*. The address of both honorary secretaries is, the British Conference on Automation and Computation, c/o Institution of Electrical Engineering, Savoy Place, London, W.C.2.

Post Office Engineering-Apprenticeship Scheme

IT was announced in Parliament by Miss Mervyn Pike, Assistant Postmaster General, that the Post Office is engaged in an expanding nation-wide programme of mechanization and automation. To enable it to carry this through, men will be required to train for key engineering-posts. A scheme is being launched under which university training or a diploma in technology course at a college of advanced technology will be guaranteed to about twenty students. During the student apprenticeship, including the time of attendance at university or college, salaries will be paid according to age, and varying from £270 per annum at seventeen to £430 at twenty-one and over. Student apprentices will be selected from young men of British nationality who are between the ages of seventeen and twenty on June 1 in the year when apprenticeship starts. The university or college studies will start in September/October of the year after apprenticeship begins. The student apprenticeship will last 4½ years, except in Scotland and Northern Ireland, where it will be slightly longer to allow for the duration of university courses. Further information can be obtained from the Engineering Chief (O.51), Post Office Engineering Department, 2–12 Gresham Street, London, E.C.2. Applications for 1960 must be submitted by February 26.

National Science Foundation : Science Faculty and Senior Postdoctoral Fellowship Awards for 1960

THE National Science Foundation has awarded 285 Science Faculty Fellowships in the sciences for the fiscal year 1960. These fellowships are offered as a means of improving the teaching of science, mathematics and engineering in American colleges and universities, and their primary purpose is to provide an opportunity for college and university science teachers to improve their work as teachers. Candidates must have a first degree or its equivalent, demonstrated ability and special aptitude for science teaching and advanced training, and three years of full-time science teaching at the college-level. There were 762 applicants from all parts of the United States and its territories.

The Foundation has also announced the award of 75 Senior Postdoctoral Fellowships in the sciences for the fiscal year 1960, selected from 259 applicants. These Fellowships were awarded to scientists of demonstrated ability and special aptitude for productive scholarship in the sciences. Twenty-eight awards were made in the life sciences, and forty-seven in the physical sciences.

High Mach Number Air-breathing Machines

THE Combustion and Propulsion Panel of the Advisory Group for Aeronautical Research and Development, North Atlantic Treaty Organization, will hold its fourth colloquium in Milan during April 4–9. The meeting will be devoted to discussions on "High Mach Number Air-breathing Engines". The opening session will consist of a round-table discussion on "The Future of Air-breathing Engines". Invited papers from Germany, Italy, the United Kingdom and the United States will deal with: (1) performance and applications, (2) hypersonic inlet studies, (3) supersonic combustion and detonation, (4) nozzle flow with chemical reactions, (5) rotating machinery, and (6) high-temperature material problems. Although the meeting is unclassified from a security point of view, attendance is limited to those who are authorized to attend by either the Group national delegates, or by members of the Combustion and Propulsion Panel.

Ford Foundation Grant to Cern

THE European Organization for Nuclear Research has received from the Ford Foundation a grant amounting to five hundred thousand dollars to cover a period of four years The facilities available at Cern now consist of two big particle accelerators, a 600 MeV. synchrocyclotron and a 28,000 MeV. proton synchrotron. The latter, the most powerful of its type in the world, has recently started operating with outstanding success. This grant follows a previous one of four hundred thousand dollars contributed by the Foundation in 1956 to cover similar expenses during four years.

Announcements

IT is announced that Cornelius Lanczos, senior professor in the School of Theoretical Physics at Dublin Institute for Advanced Studies, has recently been awarded the Chauvenet Prize by the Mathematical Association of America for an article entitled "General Theory of Linear Differential Operators". This article is to form the last chapter of a book which is to be published by Van Nostrand, entitled "Linear Differential Operators", by Prof. Lanczos.

MR. P. N. HARVEY has been appointed farm director at Gleadthorpe Experimental Husbandry Farm, Notts., of the Ministry of Agriculture, Fisheries and Food, in succession to Mr. P. J. Jones, who has been transferred to Bridget's Experimental Husbandry Farm.

PROF. MICHAEL POLANYI, emeritus professor of social studies in the University of Manchester, will give the 1960 Eddington Memorial Lecture in Cambridge at the Arts School Lecture Theatre, Bene't Street, on February 16, at 5 p.m. The title of his lecture is "Beyond Nihilism".

THE McCrone Associates of the United States are sponsoring a National Microscopy Symposium which will be held at the Del Prado Hotel in Chicago during June 13–15. The Symposium will cover (1) Instrumentation, (2) Methods and Techniques, and (3) Applications. Further information can be obtained from Walter C. McCrone Associates, 501 East 32nd Street, Chicago 16, Illinois.

Nos. 7, 8 and 9 of the Bibliography of Scientific Publications of South and South-East Asia, for July, August and September 1959, respectively, compiled jointly by Unesco Science Co-operation Offices for South and South-East Asia and published by Insdoc, National Physical Laboratory, New Delhi, contain a further 1,660 classified entries, together with author index, and the August issue includes also the Index to Vol 4 (1958).

ERRATUM. In the communication entitled "Isolation of 2-Aminoethane Phosphonic Acid from Rumen Protozoa", by M. Horiguchi and M. Kandatsu, in *Nature* of September 19, p. 901, Fig. 1 has unfortunately been inverted.

THE WILDFOWL TRUST

THE report* of the Wildfowl Trust for 1957–58 describes the administrative activities of the staff, the state of the accounts (satisfactory), the growth in membership (from 5,094 in 1957 to 5,320 in 1958), the work of scientific members of staff during the year, as well as investigations and observations of wildfowl by many well-known correspondents throughout the world. One of the latter is an interesting account by Eric Fabricius, of the University of Stockholm, of the mechanisms which make plumage waterproof. Fabricius found that ducklings of several species lost their waterproofing on a diet consisting of raw crushed fish, and regained it on a diet of insects; even in specimens that had lost their waterproofing there were indications that the oil gland was functioning.

The oil gland was removed from fourteen young tufted ducks just after hatching, while their down feathers were still enclosed in their isolating horny sheaths. After bursting of these sheaths and the full development of the down, the plumage was as repellent to water as that of the normal control birds, indicating that it is not the secretion of the oil gland which makes the plumage waterproof.

Operated and normal control birds alike lost their waterproofing when subjected to contact with smearing substances. In this condition the barbules of the down feathers were disarranged, sticking together in groups, between which were irregular spaces. The normal waterproofing was gradually restored if the birds were allowed to preen. Plumage remains water-repellent when birds feed on insects because insects are dry and clean. Plumage even remains water-repellent on a diet of fish, provided that the pieces of fish are only given to the birds while these are swimming in clean water. The preening consists of a number of stereotyped movements. The nibbling type of preening movements, in particular, aid in maintaining the barbules in a state which makes the plumage repellent to water. These findings confirm the view of Marsden that the large amount of finely distributed air among the ramifications of the feathers is the principal factor in the water-repellency of plumage.

The causes of death of birds at Slimbridge during 1955–57 have been analysed by A. R. Jennings, of the University of Cambridge. Of the 680 birds examined, the largest number of deaths were due to the high incidence of acute pulmonary congestion and pneumonia among very young birds. Sudden lowering of the air temperature often associated with heavy rainfall brings about this pulmonary syndrome, which is often referred to as 'chilling'. This type of acute respiratory disease is a common cause of high mortality in free-living species, and the same conditions seem to apply even under conditions of semi-captivity. The changes at post-mortem are quite typical and comprise acute congestion and œdema of the lungs, an enlarged gall bladder full of viscous bile and usually an unabsorbed yolk sac. In all, 138 birds died from various infections. Aspergillosis was the most frequent with 117 cases, the predilection sites for the fungus being the lungs and air sacs. The lesions were usually typical fungal granulomata, but in some birds there was an acute diffuse pneumonia. Tuberculosis was the next most common disease, and eight cases were seen.

One of the main objectives of the research programme at Slimbridge has been to obtain a better understanding of the taxonomy of the Anatidæ. All the work has now been surveyed by Dr. G. U. T. Matthews, the assistant director of the Trust, who shows that a whole variety of techniques are open to the modern taxonomist. Many of these techniques are incapable of assessing relationships below the family level and are only of passing interest when concerned with relationships within a family. It is beginning to appear that the Anatidæ is an unusually homogeneous and closely knit family.

What makes the report outstanding, of course, is the revealed and hidden imprint of Peter Scott, the director. His sketches lose none of their attractiveness with the years; they may even excite more interest as they become more audacious. The many photographs are included only if they illustrate the work of the Trust and do so with adornment; not one of them is below standard. With a report of such high quality it is to be hoped that the activities of the Inland Revenue authorities may not, as is feared, lead to the production of future reports in an attenuated form.

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* Tenth Annual Report of the Wildfowl Trust, 1957–1958. Edited by Peter Scott and Hugh Boyd. Pp. 184+32 plates. (Slimbridge, Glos.: The Wildfowl Trust, 1959.) 10*s*. net.

DAIRY RESEARCH IN SCOTLAND

THE recent triennial report of the Hannah Dairy Research Institute* gives a clear picture of the expansion of the Institute's activities in the past three years. New laboratories have been built, and greatly increased facilities have been added to those formerly available for metabolic studies. The work reported continues to reflect the aims defined when the Institute was founded some thirty years ago.

Among several important additions to the Institute is a splendidly equipped climatic laboratory. This has been constructed, with capital provided by the Colonial Office, and is being used for studies on the effect of climate on the physiology of cattle. The laboratory includes a room in which a cow or steer can be maintained at any temperature from 0° to 60° C. within ± 0·1° C., and at a relative humidity within ± 1 per cent over a wide range. A second room houses four cattle with slightly less critical control of environment. Metabolism houses have also been constructed for sheep and cattle. These contain six new closed-circuit respiration chambers for sheep and one for cattle, and provision is also made for twenty-four sheep metabolism cages. With their unique equipment and instrumentation the climatic and metabolism buildings are a notable example of collaboration between biologists, physicists and engineers.

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* Hannah Dairy Research Institute. Report for the Three Years ended March 31, 1959. Pp. 52.

The Grass and Dairy Husbandry Department has continued its investigation of the intensive management of a largely grassland dairy farm. Purchases of fertilizer rather than concentrates continue to show a high return, and the Ayrshire herd averaged 800 gallons a year with one cow per 1·8 acre. The choice of forage species and strains, coupled with suitable management and fertilizer treatments, is being studied in extensive plot and feeding experiments. In the Biochemistry Department fundamental studies are in progress concerning the chemistry of milk protein and the vitamins and enzymes of milk. Attention has been given to factors influencing the important riboflavin and vitamin A components of milk, to the synthesis of lysine in the rumen and to the effect of thyrotropin and growth hormone on milk yield.

The construction of new equipment has not prevented the Nutrition Department from continuing its important studies on energy metabolism. These have shown that above the maintenance-level the utilization of mixtures of volatile fatty acids by sheep decreases with decreasing chain-length, being as low as 30 per cent with some mixtures rich in acetic acid. In ruminants almost all dietary carbohydrate is absorbed as these short-chain acids, and world-wide interest has been aroused by this first step in the reconciliation of classical energy studies with recent developments in ruminant nutrition.

Other work with sheep has been concerned with the effects of environmental temperature and clipped fleeces on energy metabolism, the influence of the physical state of roughages on their net energy and the role of vitamin E in muscular disease. Cow-feeding experiments suggested that the allowance of concentrates per gallon of milk should increase at higher yields.

The Physiology Department is concerned with the physiological responses of cattle to climatic stress. While aimed at the improvement of cattle productivity in hot countries, this work must also provide fundamental information of value in studies on housing in Britain. Two small chambers have been used for investigating the physiology of panting; the thermal stimulus to panting was found to be effective, in calves, only on the skin or upper respiratory tract. The ability to reduce tidal volume with increasing thermal stress appeared to be associated with high heat-tolerance as indicated by rectal temperature.

The validity of rectal as a measure of deep body temperature, the effects of heat stress on the blood and the role of the skin in loss of heat were also investigated. Important studies on the rate of acclimatization of calves to heat are now being extended to cows.

The Department of Veterinary Pathology studies both practical and applied aspects of mastitis. Histological examination of udders from infected cows, the incidence on farms of different types of mastitis and the relationship between the Whiteside test and cell counts have all been considered.

The Bacteriology Department has been largely concerned with the important *coli-aerogenes* bacteria of milk and with the application of the coliform test. Demand for sterilized milk in cities has prompted investigation of the spore-forming bacteria of such milk and of tests for effective 'sterilization'.

Many practical aspects are also dealt with by the Department of Technical Chemistry. Welcome collaborative studies are reported on the accuracy of various analytical methods for milk, and these have led to revision of the formula for the calculation of solids-not-fat content by means of the lactometer. Feeding trials indicated that with spring grazing the usual increase in milk protein is probably due more to an improved plane of nutrition than to an increase in any particular component of grass. Long-term studies have dealt with factors influencing the coagulation of casein, a problem in the manufacture of dairy products, and with the production, preservation and storage of dried milk. A new system was developed for the fractionation of the carbohydrates of roughages.

Readers of this report are provided with a ready reference to more detailed information by the liberal inclusion of relevant reprint numbers.

C. C. BALCH

CORRELATION OF AUDIO-FREQUENCY ELECTROMAGNETIC RADIATION WITH AURORAL ZONE MICROPULSATIONS

By DR. J. AARONS
Electronics Research Directorate, Air Force Cambridge Research Center
AND
G. GUSTAFSSON and A. EGELAND
Kiruna Geophysical Observatory

DURING three periods of approximately one month each, continuous recordings were taken of the electromagnetic energy in the spectrum region of 10 c.p.s. to 10 kc.p.s. The detection equipment was a 10-m. vertical antenna, a swept frequency battery-operated spectrum analyser and a recording milliammeter. The frequency analyser had two ranges; it first swept through its lower range of 10–1,000 c.p.s., with a 1 c.p.s. band-width. A second sweep was then initiated to go from 500 c.p.s. to 10 kc.p.s. with a 40-c.p.s. band-width. A total time for a complete recording of both of the sweeps was 32·8 min. The equipment was housed at an isolated area about 2 km. from the nearest 50 c.p.s. power line. An example of the registration of the higher-frequency range is given in Fig. 1.

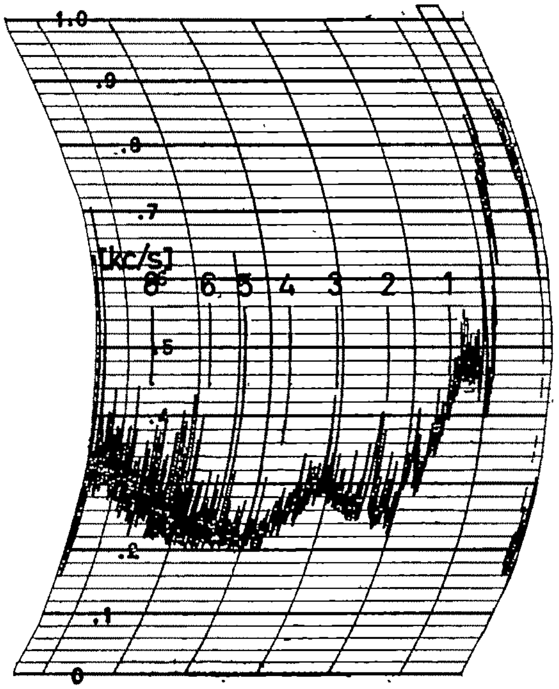

Fig. 1. Recording of the electromagnetic radiation of December 5, 1828 to 1839 M.E.T., 1958, in the frequency range 0·5–10 kc./s.

The aim of the programme carried out near the Kiruna Geophysical Observatory, Sweden (geographic co-ord., 67·8° N., 20·4° E.; geomagnetic co-ord., 65·3° N., 115·5° E.), was twofold, namely: (1) to investigate changes in the background signal-level; (2) detect emission or absorption of energy at the gyro-frequency of solar terrestrial ions.

Several magnetically disturbed periods occurred during the measurement periods October 12–November 11, 1958, December 12, 1958–January 11, 1959, and April 15–May 14, 1959. On eleven occasions electromagnetic radiations associated with micropulsations of the Earth's magnetic field were detected. Two distinct frequency-bands were identified in this low-frequency emission. The lower-frequency emission, in the range 0·5–1·4 kc./s., has been mentioned by Egeland[1] as "noise" during aurora, while the higher-frequency emission, in the range 1·8–4·5 kc./s., was identified by Watts[2] as "hiss". For the lower-frequency emission very little results have hitherto been obtained.

Fig. 2 is a spectrum analysis of the signal received on December 5, 1958. For 13 hr. from 0701 to 2010 M.E.T. a distinct signal was seen in one or both of the two bands. Since the results are recorded in the form of a spectrogram, only the frequency variations are shown here. The cross at the centre of each sweep denotes the frequency of maximum intensity, while the length of line indicates the frequency-width of the emission above that of the background signals. For these sweeps the 'hiss' signals maxima varied from 2·3 to 3·4 kc./s. By contrast, the lower-frequency emissions do not vary more than + or − 150 c.p.s. around 750 c.p.s. The band-width of the lower-frequency emissions is less than that of the upper band, as can be seen on the spectrograms in Fig. 2. The error of reading and the band-width of the equipment lead to a value of + or − 100 c.p.s. for the accuracy of the measurements. Thus, while the lower-frequency band remained fixed in frequency the 'hiss' emissions varied. The amplitudes of each of the bands fluctuated from sweep to sweep during every disturbed period.

In all the eleven observed periods of emissions, micropulsations of the magnetic field were shown on the Kiruna magnetograms. The micropulsations seen in the auroral zone show periods of the order of 0·5–5 min. and maximum displacement of 50γ. These do not occur during the largest deflexions of the magnetic storms. In four of the longer periods of radiation detected, the low-frequency emissions occurred between two phases of a magnetic storm when micropulsations were present.

The Kiruna magnetogram for December 5 is shown in Fig. 3. A tracing of the outlines of six consecutive recordings during the December period is shown in Fig. 4. The time of duration of each tracing is about 10 min. The low-frequency tracing is not included, and therefore there is a gap of 23 min. between the recordings. The dotted line is the no-signal receiver noise line. The curvature of the zero line is due to equipment parameters. The vertical amplitude scale is logarithmic. The signals are at best of extremely low amplitude, rarely exceeding 5 μV. from the antenna.

The 'hiss' signals received have been observed before, but at lower latitudes. Watts[2] observed 'hiss' for 12 hr. at a site near Boulder, Colorado. The duration of our longest period recorded is somewhat longer than this. The presence of so many examples of this low-frequency emission during the short period of recordings indicates its prevalence in the auroral zone during periods of magnetic activity of the micropulsation types.

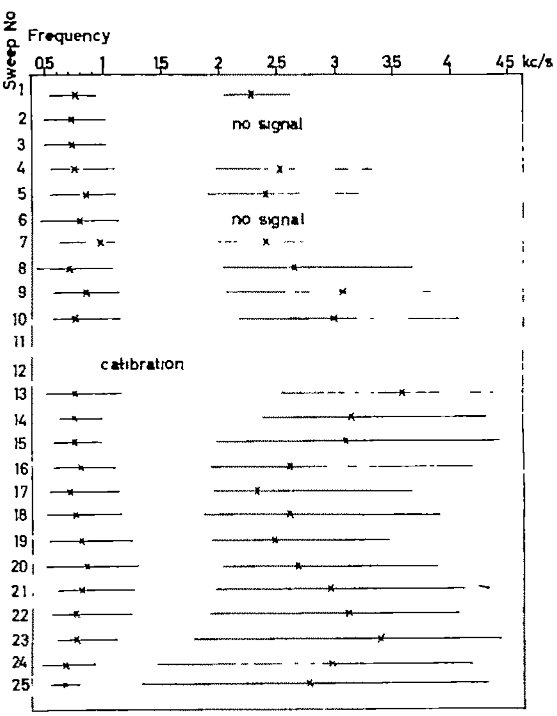

Fig. 2. A spectrogram of the signal received on December 5, 0701 to 2010 M.E.T., 1958. The length of line indicates the frequency-width. The cross on each line denotes the frequency of maximum intensity

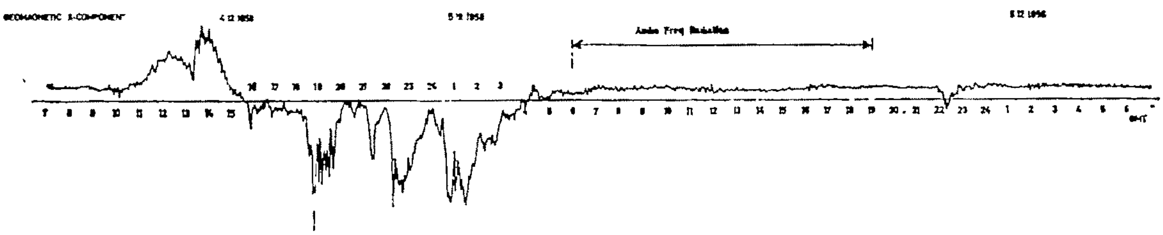

Fig. 3. The horizontal component of the geomagnetic field at Kiruna

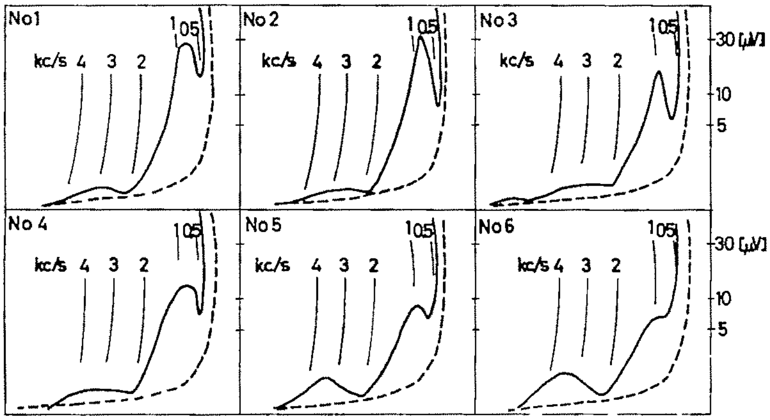

Fig. 4. Tracing of the outlines of six consecutive recordings of the December period 1958

The lower-frequency sweep has not as high a sensitivity as the higher one. The results from this channel indicate, however, that the 750 c.p.s. emission is relatively wide in spectrum. A sharply defined signal would have increased the level considerably in the lower-frequency sweep. It increased it only slightly.

The origin of low-frequency emissions has been discussed by various authors who have widely differing ideas. Ellis[3] states that Čerenkov radiation is the probable cause of 4–5 kc./s. auroral signals, Gallet[4] and Gallet and Helliwell[5] base their theory on an analogy to travelling-wave-tube amplifiers with modifications to suit the solar–terrestrial parameters and Aarons[6] compares the gas discharge mechanism with the aurora. Discussions of the possibility of detecting the protons and the other ions by the emissions is also discussed by Aarons.

One reason for being able to detect the signal is the low background-level of noise at the remote site and the low signal-level from atmospherics at Kiruna. Analysis of the three-months period (to be described in another publication) indicates that the normal diurnal variations of the atmospherics were not correlated with magnetic disturbances described in this article.

The lower-frequency emissions are of greater interest than the upper band principally because they lie for the most part near 750 c.p.s., the gyro-frequency of protons in the Kiruna area at an altitude of 100 km. Table 1 shows the gyro-frequency for protons in Kiruna as a function of altitude, calculated from the formula:

$$f_H = \frac{eH}{2\,\pi mc}$$

Fig. 2 demonstrates the stability of the lower-frequency emissions. However, on May 5, 1959, an emission band centred at 1 kc./s. was detected. In subsequent sweeps the single band began to split into two bands with the lower one slightly above 1 kc./s. and the upper band moving to 2 kc./s. Tracings of the signal are seen in Fig. 5. If the May 5 case is another example of 'hiss' developing into two bands it does not mitigate the case for proton radiation. However, it may well be that the lower-frequency band is a component of the 'hiss'. Only additional experimental evidence will determine this. The largest part of the data shows signals near 750 c.p.s. with relatively small changes in frequency.

Table 1

Altitude	Kiruna
0 km.	794 c./s.
60 km.	771 c./s.
100 km.	755 c /s.
200 km.	718 c./s.

The evidence in this article is rather clear. There are excellent correlations between the auroral zone micropulsations and the radiation in the audio-frequency band. Two distinct frequency bands have appeared on 12 per cent of the days for which results were obtained. This is a lower limit since on several days when the sensitivity was low, emissions were not detected. The upper-frequency band is 'hiss'. It varies in frequency throughout the recording period.

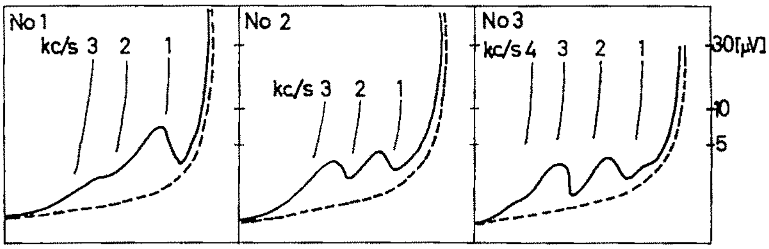

Fig. 5. Tracing of the outlines of three consecutive recordings of the May period 1959

The lower band at 750 c.p.s. for almost all the cases is steadier in frequency. Its spectral location indicates that it may well be radiation from protons, but its occasional change in frequency may make it another component of the 'hiss' signals.

We are indebted to Dr. B. Hultqvist, director of the Kiruna Geophysical Observatory, for his interest and advice.

The programme was in part supported by the U.S. Air Research and Development Command (Contract 61(514)-(1314)).

[1] Egeland, A., Institute of Theoretical Astrophysics, Blindern, Oslo, Report No. 6 (1959).

[2] Watts, J. M., *J. Geophys. Res.*, **62**, 199 (1957).

[3] Ellis, G. R., *J. Atm. Terr. Phys.*, **10**, 302 (1957).

[4] Gallet, R. M., *Proc. Inst. Radio Eng.*, **47**, 211 (1959).

[5] Gallet, R. M., and Helliwell, R. A., *J. Res. N.B.S.*, **63**, *D*, 21 (1959).

[6] Aarons, J., doctoral thesis, University of Paris (1954).

CELL-GROWTH OF PLANT ROOTS, INORGANIC SALTS AND NATIVE SERUM PROTEINS

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THIS article deals with the influence of increasing concentrations of some inorganic salts on the phytohormonal activity of native human serum compared with the influence of indolyl-3-acetic acid, which is considered the most important phytohormonally active substance in serum[1].

The phytohormonal activity of human serum, that is, its inhibitory effect on the growth of *Lupinus albus* roots, is influenced by the salt concentration in the nutrient solution with which the serum is diluted. Thus, the inhibitory effect of serum when diluted with distilled water is far less than when the serum is diluted with a plant-physiologically well-balanced triple-salt solution consisting of calcium nitrate 2·5 m*M*, magnesium sulphate 7·5 m*M*, and potassium dihydrogen phosphate 9·0 m*M* (after Shive[2])[3]. The increased effect of serum in a solution of salts, as the result of single-salt experiments, is interpreted as a phytohormone-separating effect of, especially, the calcium ions on the serum proteins[4].

The phytohormonal effect of serum is diminished, however, when the concentration of inorganic salts in a serum-salt solution is increased further, as will be accounted for below.

Fig. 1 shows the results of four, mutually independent, experiments (I, II, III and IV) with the growth of *Lupinus albus* roots in nutrient solutions, partly with and partly without admixture of native serum at the concentrations 3, 2, 2 and 1 per cent (v/v). The nutrient solutions used with or without the addition of serum comprise distilled water, Shive's solution, and solutions of the individual salts contained in Shive's solution. 1/1 × Shive thus denotes the salt solution specified above. 2/1, 3/1 and 5/1 × Shive denote salt solutions in which all the salt components are present at concentrations 2, 3 and 5 times that of 1/1 × Shive's solution. The upper curves of Fig. 1 show the growth in solutions without serum; the lower curves show the growth in solutions with serum.

The osmotic pressure in Shive's solution is about 1·3 atmospheres. Consequently, 5 × Shive has an osmotic pressure of about 6·5 atmospheres, corresponding to the osmotic pressure of undiluted human serum. It is a familiar fact that the osmotic pressure of serum proteins is extremely low: about 0·033 atmospheres.

In Fig. 1 the salt concentrations are shown on the axis of abscissæ, while the axis of ordinates shows the elongation of the roots in mm. per 24 hr. at 15° C. (the Macht–Livingston technique[5]). The salt concentration 0 in experiments I and II indicates that in these experiments distilled water has been used,

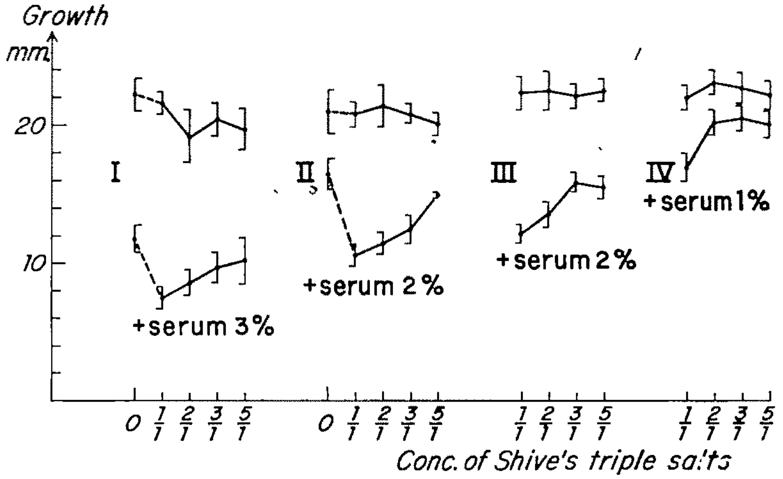

Fig. 1 Experiments I, II, III and IV. The phytohormonal effect, that is, the root-growth-inhibitory effect, of native, human serum at concentrations of 3, 2, 2 and 1 per cent (lower curves). For comparison, the upper curves show growth in the same solutions without serum, that is, in distilled water (experiments I and II only), in Shive's triple-salt solution, and in solutions with all the Shive salts multiplied by 2, 3 or 5 (experiments I to IV). Axis of ordinates: growth of roots in mm. in 24 hr. at 15° C.

partly with and partly without admixture of serum as nutrient solution.

The upper curves of Fig. 1 present the elongation in the above-named salt solutions and in distilled water without serum, respectively. It will be seen that in all solutions with serum the growth is unaffected irrespective of whether the salt concentration is one or five times the concentration in Shive's solution. In 24-hr. experiments the elongation in distilled water (the first two values on the upper curves for experiments I and II) is 0–10 per cent greater than in Shive's solutions[5,6].

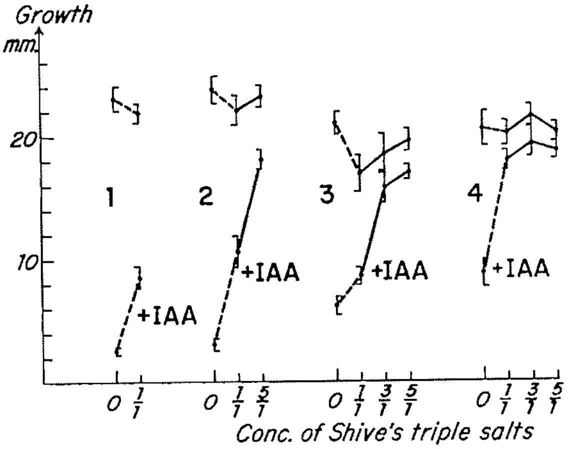

Fig. 2. Experiments 1, 2, 3 and 4. The phytohormonal effect, that is, the root-growth inhibitory effect, of indolyl-3-acetic acid (IAA) 10^{-7} *M* (lower curves). Upper curves show comparative growth in the same solutions without indole-3-acetic acid, that is, in distilled water, in Shive's triple-salt solution, and in triple-salt solutions with all the Shive salts at concentrations multiplied by 3 or 5. The concentration in Shive's solution alone is denoted by 1/1. Axis of ordinates: growth of roots in mm. in 24 hr. at 15° C.

From the first two values on the lower curves, representing the results of experiments I and II, it will be seen that in the solutions containing serum diluted with distilled water the elongation is greater than in the serum solutions diluted with Shive's salts. The growth depression brought about by the admixture of Shive's salt to the serum solution is accounted for in detail in a previous paper[4], and as mentioned, it is due to a specific serum-protein/inorganic-salt interaction.

In the solutions containing serum diluted with the various Shive salts, represented by the four lower curves in Fig. 1, which give the results of experiments I–IV, it will be seen that the elongation increases with a salt concentration increasing beyond the concentration in 1/1 × Shive. In all the experiments the elongation in the serum solutions containing Shive salts is thus lowest when the solution is diluted with 1/1 × Shive.

How, then, is this reduced phytohormonal activity of serum with increasing inorganic ion concentration to be interpreted ?

It is impossible to give an exhaustive explanation of this phenomenon ; but a partial explanation may be found by comparing the effect of increasing salt concentrations on a pure, unbound phytohormone in a corresponding experimental series and the effect just named of the same salts on the protein-bound phytohormone in human serum.

Fig. 2 serves this purpose. It shows the results of four experiments (1, 2, 3 and 4), in which indolyl-3-acetic acid is dissolved, partly in distilled water, partly in Shive's solution, and partly in solutions containing increasing concentrations of the individual Shive salts, as described previously for Fig. 1. From the upper curves for the four experiments in Fig. 2 it will be seen that, as was the case in Fig. 1, the elongation in the salt concentrations without indolyl-3-acetic acid is, in all essentials, constant with increasing salt concentration (experiment 3 of Fig. 2, however, shows a slight increase with increasing salt concentration), while, as previously, the elongation in distilled water with indolyl-3-acetic acid is greater than, or of the same order as, the elongation in the salt solutions.

From all the lower curves in Fig. 2 it will be seen that the root-growth inhibition, the phytohormonal activity, of the unbound indolyl-3-acetic acid in an aqueous solution at the concentration 10^{-7} *M* is greatly pronounced but decreases rapidly when the indolyl-3-acetic acid solution contains a certain amount of inorganic salts (with calcium ions). This effect is not known to have been described before for the root-growth experiments, and it is preponderantly due to the presence of calcium ions in the solutions[4]. For *Avena* coleoptiles, Thimann and Schneider[7] have described a similar calcium–auxin antagonism in single-salt experiments with calcium chloride. The increased auxin-induced elongation of coleoptiles was thus inhibited by the addition of calcium at concentrations above 0 001 *M*. Through experiments, likewise with *Avena*, Cooil and Bonner[8] demonstrated that this calcium effect is associated with a tightening effect upon the cell wall and is apparently brought about through interference with the normal processes involving auxin.

Like coleoptile cells, also plant root cells become apparently more resistant to action of externally applied phytohormones with increasing calcium concentrations in the nutrient solution. The effect of increasing concentrations of the Shive salts beyond the concentration in 1/1 × Shive in the serum dilutions, illustrated in Fig. 1, may therefore be interpreted as being due to such an increased resistance of the roots.

Fig. 1 thus shows two salt or calcium effects, namely : (1) a primarily acting salt or calcium effect, acting on the serum proteins[3] and becoming apparent by a comparison of the phytohormonal effect (root-growth inhibition) of serum diluted with distilled water and the phytohormonal effect of serum diluted with 1/1 × Shive's triple-salt solution ; (2) a secondarily acting salt or calcium effect, acting on the plant root cells and becoming apparent by an additional supply of Shive salts or calcium ions to the serum solution beyond the concentration in 1/1 × Shive.

This work was supported by grants from P. Carl Petersen's Foundation, the Foundation of Johan Weimann, Merchant, of Odense, and Hanne Weimann, *née* Seedorff, and King Christian X's Foundation.

[1] Rennfke, F., *Acta Pharmacol. et Toxicol.* (in the press).
[2] Shive, J. M., *Amer. J. Bot.*, 2, 157 (1915).
[3] Rennfke, F., *Nature* (in the press).
[4] Rennike, F., *Acta Pharmacol. et Toxicol.*, 16, 97 (1959).
[5] Rennike, F., *Physiol. Plant.*, 11, 421 (1958).
[6] Ekdahl, I., *Ann. Roy. Agric. Coll. Sweden*, 23, 497 (1957).
[7] Thimann, K. V., and Schneider, C. L., *Amer. J. Bot.*, 25, 270 (1938).
[8] Cooil, B. J., and Bonner, J., *Planta*, 48, 696 (1957).

MICROBIAL OXIDATION OF GLYCOLLATE VIA A DICARBOXYLIC ACID CYCLE

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Medical Research Council Cell Metabolism Research Unit, Department of Biochemistry, University of Oxford

ALTHOUGH the tricarboxylic acid cycle[1] has been established as the major route for the oxidation of acetate, and of compounds catabolized to acetate, in micro-organisms as well as in other biological materials (for review, see Krebs[2,3], Elsden[4], Ajl[5] and Kornberg[6]), little is known of the pathways of oxidation of C_2-compounds more highly oxidized than acetate (such as glycollate, glyoxylate or glycine). Studies with mammals[7], with mammalian tissue preparations[8-10] and with a strain of *Pseudomonas*[11,12] have led to the suggestion that glycollate or glycine is oxidized via glyoxylate and formate (1):

$$\left.\begin{matrix}CH_2.OH.COOH\\ \text{or}\\ CH_2NH_2.COOH\end{matrix}\right. \xrightarrow{+O_2} CO_2 + HCOOH \xrightarrow{+\frac{1}{2}O_2} CO_2 + H_2O \qquad (1)$$

Evidence for the occurrence of this pathway in *Pseudomonas* rests largely on the demonstration that formate, which is rapidly oxidized, accumulates[11,12] when cells oxidizing glycine or glyoxylate are poisoned with hypophosphite[13].

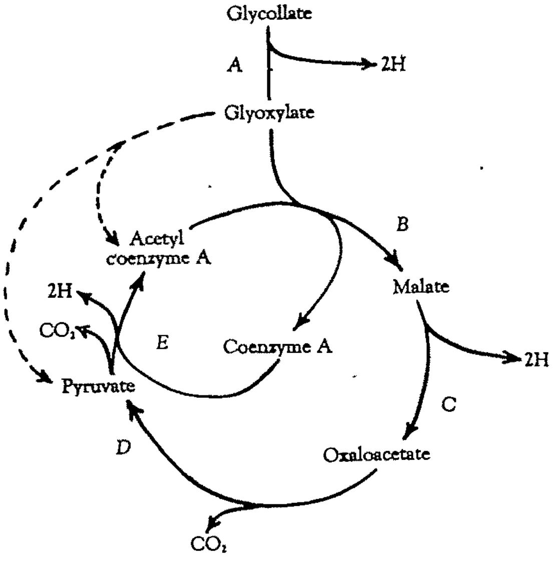

Fig. 1. Postulated dicarboxylic acid cycle for the oxidation of glycollate and glyoxylate. (The sequence of reactions is catalysed by *A*: glycollic oxidase (ref. 18), *B*. malate synthetase (ref. 19); *C*: malic dehydrogenase (ref. 20); *D*: oxaloacetic carboxylase (ref. 25) and *E*: the pyruvate oxidase system (ref 26). Each turn of the cycle results in the oxidation of one molecule of glyoxylate: $CHO.COOH + O_2 \rightarrow 2CO_2 + H_2O$. The dotted line represents the pathways from glyoxylate to cycle intermediates, first shown by studies with *Ps. ovalis* Chester (ref. 18), which also operate in the strain of *E. coli* used in the present work, and which are a means of providing for the net formation of precursors of cell constituents during growth)

However, the oxidation of glycollate and of glyoxylate by washed suspensions or cell-free extracts of *Pseudomonas aeruginosa*[14], of *Pseudomonas ovalis* Chester[15], or of *Escherichia coli* (strain *w*) proceeds at identical rates and to identical extents in the presence or absence of hypophosphite, although formate oxidation is completely abolished by this inhibitor. This indicates that a sequence of reactions (1) involving free formate is probably not of major importance in glycollate oxidation in these micro-organisms.

The observed effects on glycollate metabolism of other metabolic poisons, particularly of fluoroacetate[16,17], suggest that the metabolism of glycollate (and of glyoxylate derived therefrom) is linked to that of intermediates of the tricarboxylic acid cycle. One such link has been demonstrated[18]: extracts of glycollate-grown *Pseudomonas ovalis* Chester can catalyse the formation of intermediates of the tricarboxylic acid cycle from glycollate or glyoxylate. Acetyl coenzyme *A* can be formed by a sequence of reactions the overall effect of which is:

$$2 \text{ glyoxylate} + \text{coenzyme } A \rightarrow \text{acetyl coenzyme } A + 2CO_2 + H_2O \qquad (2)$$

and can react with glyoxylate to form malate (3), catalysed by malate synthetase[19]:

$$\text{glyoxylate} + \text{acetyl coenzyme } A + H_2O \rightarrow \text{malate} + \text{coenzyme } A \qquad (3)$$

Malate can be oxidized to oxaloacetate[20], which can condense[21] with acetyl coenzyme *A* formed in reaction (2) to yield citrate. Since citrate can be metabolized further through the tricarboxylic acid cycle, this sequence of reactions might serve not only to provide carbon skeletons for the biosynthesis of cell constituents when micro-organisms grow on C_2-compounds more highly oxidized than acetate[18,22,23], but might also serve as part of the terminal pathway of oxidation of such C_2-compounds.

This point was tested with a mutant of *Escherichia coli*[24] devoid of the citrate-forming condensing enzyme[21] and hence unable to utilize acetate and unable to form citrate and glutamate. The results obtained show that although intermediates of the tricarboxylic acid cycle are required for growth, this cycle is not an obligatory pathway for the oxidation of glycollate. Glycollate oxidation can be effected

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via a dicarboxylic acid cycle (Fig. 1), which obviates the necessity for reduction of glyoxylate to the oxidation level of acetate prior to its catabolism. It brings about the oxidation of glyoxylate (derived from glycollate[18], Fig. 1*A*) by condensation with acetyl coenzyme *A* to form malate[19] (reaction 3; Fig. 1*B*); oxidation of malate to oxaloacetate[20] (Fig. 1*C*); formation of carbon dioxide and pyruvate from oxaloacetate, possibly with intermediary formation of phospho-pyruvate[25] (Fig. 1*D*) and regeneration of the acetyl coenzyme *A* acceptor molecule from pyruvate by oxidative decarboxylation with the pyruvic oxidase system[26] (Fig. 1*E*). A major difference in principle between oxidations effected by the tricarboxylic acid cycle and those effected by the postulated dicarboxylic acid cycle is that, while in each turn of the former, one molecule of acetyl coenzyme *A* is completely oxidized with the obligatory participation of a keto-acid (which is regenerated), in each turn of the latter, one molecule of keto-acid is completely oxidized with obligatory participation of acetyl coenzyme *A* (which is regenerated).

The evidence for the cycle was obtained from studies on the growth of the *E. coli* mutant *M* 22–64 [24], on oxidations effected by washed suspensions of the organism, and on enzymic reactions catalysed by extracts obtained after disintegration of the organism in a magnetostrictor oscillator. The main findings are as follows:

(1) The mutant grew readily on a medium containing glycollate (50 m*M*) plus glutamate (2 m*M*) as carbon sources, but did not grow on glycollate alone, or on acetate either in the presence or absence of glutamate. Growth on glycollate plus glutamate was stimulated both in rate and extent by the addition of acetate (2 m*M*). These results confirm[24] that glutamate, which is required for growth, cannot be synthesized by the mutant deficient in the ability to form citrate, and they support the view that the tricarboxylic acid cycle operates primarily to supply cell constituents under these conditions. Stimulation of growth by acetate indicated a point of entry into metabolic pathways of this compound other than via the condensing enzyme[21].

(2) Suspensions of washed cells oxidized glycollate plus acetate ($Qo_2 = 158$ µl. of oxygen absorbed/hr./mgm. dry wt.) more rapidly than glycollate alone ($Qo_2 = 109$), although they did not oxidize acetate alone. The addition of acetate, pyruvate or malate to cells oxidizing glycollate increased the quantities of oxygen absorbed from 45–59 to 83–93 per cent of that required for complete oxidation of glycollate. Pyruvate or malate were oxidized incompletely and the quantities of oxygen absorbed were consistent with their oxidation only to the level of acetate. The oxidation of glycollate was virtually complete in the presence of 2:4-dinitrophenol (1·3 m*M*).

(3) Washed suspensions of the mutant incorporated isotope from added [1-^{14}C] or [2-^{14}C]glycollate initially into malate, and thereafter into aspartate, alanine, phosphorylated compounds and sugars. No radioactivity was detected in glutamate, citrate or other intermediates of the tricarboxylic acid cycle.

(4) The addition of [2-^{14}C]acetate to cells oxidizing unlabelled glycollate resulted in the rapid appearance of the isotope in oxaloacetate and pyruvate. Similar results were obtained with [2-^{14}C]glycollate; labelled oxaloacetate and pyruvate accumulated in the presence of arsenite (10 m*M*).

(5) Washed suspensions of the mutant, shaken in air at 30°, did not evolve labelled carbon dioxide from [1-^{14}C] or [2-^{14}C]acetate alone, but did so rapidly if unlabelled glycollate or glyoxylate was added to the system. Malate or pyruvate was ineffective in promoting this oxidation of acetate.

(6) The rates and extent of evolution of labelled carbon dioxide from malate, labelled with carbon-14 in the carboxyl groups[27], were stimulated by the presence of glycollate or glyoxylate. This stimulation was abolished by arsenite (10 m*M*). Labelled oxaloacetate and pyruvate were formed in these reactions. Pyruvate or acetate was ineffective in stimulating release of labelled carbon dioxide.

(7) Cell-free extracts of the mutant readily catalysed the formation of malate from acetyl coenzyme *A* and glyoxylate, the activity of the malate synthetase[19] being exceptionally high and sufficient to effect the formation of 20 µmoles of malate/hr./mgm. of soluble protein under the conditions of assay[28]. Acetyl coenzyme *A* was not split by the extracts when oxaloacetate was added, confirming[24] the absence of the citrate-forming condensing enzyme.

(8) The activity of malic dehydrogenase[20] in such extracts was sufficient to catalyse the oxidation of 70 µmoles of reduced diphosphopyridine nucleotide/hr./mgm. of soluble protein when incubated with this coenzyme and oxaloacetate.

(9) The cell-free extracts catalysed the anaerobic decarboxylation of oxaloacetate, the initial rate of release of carbon dioxide (7·8 µmoles/hr./mgm. of soluble protein) being doubled by the presence of adenosine triphosphate[25].

(10) Although the activity of reduced diphosphopyridine nucleotide oxidases was too high to permit a quantitative assay to be made of the pyruvate oxidase system[26], the cell-free extracts formed at least 9·9 µmoles of reduced diphosphopyridine nucleotide/hr./mgm. of soluble protein when incubated with pyruvate, glutathione, magnesium chloride, thiamin pyrophosphate and coenzyme *A*, which showed that acetyl coenzyme *A* could be readily formed from pyruvate by the extract. The identity of the formed acyl thiolester with acetyl coenzyme *A* was established by chromatography of the acyl hydroxamates[29].

The enzymic rates described under (7)–(10) above are more than sufficient to account for the observed rates of glycollate oxidation (3–4 µmoles/hr./mgm. dry wt.). Coupled with the findings on the growth of the mutant (1) and the oxidation of unlabelled and isotopically labelled substrates (2)–(6), these results support the assumption that the sequence of reactions outlined in Fig. 1 occurs in the mutant of *Escherichia coli* studied, and that this oxidative dicarboxylic acid cycle represents a quantitatively important route for the microbial oxidation of carbon compounds at the oxidation-levels of glycollate or glyoxylate.

Full experimental details will be published elsewhere.

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[1] Krebs, H. A., and Johnson, W. A., *Enzymologia*, **4**, 148 (1937).
[2] Krebs, H. A., "Advances in Enzymology", **3**, 191 (1943).
[3] Krebs, H. A., and Lowenstein, J. M., in "Chemical Pathways of Metabolism", 2nd ed.; ed. by Greenberg, D. M. (Academic Press Inc., New York; in the press).
[4] Elsden, S. R., in "Special Lectures in Biochemistry 1954–1955", 17 (University College, London; Distrib. by H. K. Lewis and Co., Ltd., London).
[5] Ajl, S. J., *Physiol. Rev.*, **38**, 196 (1958).
[6] Kornberg, H. L., "Ann. Rev. Microbiol.", 13, 49 (1959).
[7] Weinhouse, S., and Friedmann, B., *J. Biol. Chem.*, **191**, 707 (1951); **197**, 733 (1952).
[8] Nakada, H. I., and Weinhouse, S., *Arch. Biochem. Biophys.*, **42**, 257 (1953).
[9] Nakada, H. I., Friedmann, B., and Weinhouse, S., *J. Biol. Chem.*, **216**, 583 (1955).
[10] Nakada, H. I., and Sund, L. P., *J. Biol. Chem.*, **233**, 8 (1958).
[11] Campbell, L. L., *J. Bacteriol.*, **68**, 598 (1954).
[12] Campbell, L. L., *J. Biol. Chem.*, **217**, 669 (1955).
[13] Takamiya, A., *J. Biochem.* (*Tokyo*), **40**, 415 (1953).
[14] Bachrach, U., *Biochem. J.*, **66**, 559 (1957).
[15] Kornberg, H. L., and Gotto, A. M., *Biochem. J.* (in preparation).
[16] Kalnitsky, G., and Barron, E. S. G., *J. Biol. Chem.*, **170**, 83 (1947).
[17] Jayasuriya, G. C. N., *Biochem. J.*, **64**, 469 (1956).
[18] Kornberg, H. L., and Gotto, A. M., *Nature*, **183**, 1791 (1959).
[19] Wong, D. T. O., and Ajl, S. J., *J. Amer. Chem. Soc.*, **78**, 3230 (1956).
[20] Mehler, A. H., Kornberg, A., Grisolia, S., and Ochoa, S., *J. Biol. Chem.*, **174**, 961 (1948).
[21] Ochoa, S., Stern, J. R., and Schneider, M. C., *J. Biol. Chem.*, **193**, 691 (1951).
[22] Callely, A. G., and Dagley, S., *Nature*, **183**, 1793 (1959).
[23] Quayle, J. R., and Keech, D. B., *Nature*, **183**, 1794 (1959).
[24] Gilvarg, C., and Davis, B. D., *J. Biol. Chem.*, **222**, 307 (1957).
[25] Utter, M. F., and Kurahashi, K., *J. Biol. Chem.*, **207**, 787 (1954).
[26] Korkes, S., in "Methods in Enzymology", ed. by Colowick, S. P., and Kaplan, N. O., 1, 490 (Academic Press Inc., New York, 1955).
[27] Dixon, G. H., Kornberg, H. L., and Lund, P., *Biochim. Biophys. Acta* (in the press).
[28] Dixon, G. H., and Kornberg, H. L., *Biochem. J.*, **72**, 3, P (1959).
[29] Stadtman, E. R., and Barker, H. A., *J. Biol. Chem.*, **184**, 769 (1950).

FUNCTIONAL PITUITARY TRANSPLANTS IN RATS

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IN a paper published a number of years ago[1] one of us presented evidence demonstrating that anterior pituitary transplants from infantile rats grafted into the anterior eye chamber of hypophysectomized adult males will often restore the functional activity of the testes. It is true that in these early experiments no histological study of the sella was undertaken, although only those animals from which the pituitary gland was removed as a whole, practically uninjured, were considered for further study.

More recently, fresh work by Courrier[2] and by Fry and Long[3] has been published indicating that the effect of the functional activity of pituitary grafts, not only on the testes but also on other end-organs, may be far greater than is attributed to them at the present time. Owing to the multiple dependence of the pituitary gland upon the hypothalamus, and to the role, recently revealed, that the former takes in the functional activity of the latter, the prevailing opinion to-day is that the anterior pituitary by itself is capable of very little of the functional activity by which this organ is characterized in its normal position (see refs. 4 and 5). Prompted by the work of Courrier and of Fry and Long, we have repeated our work on 38 adult male rats with strict observance of all control experiments including a large number of hypophysectomized controls, histological study of the sella, mating of the animals under experiment with normal females and in four cases enucleation of the eyes bearing the grafts (see Table 1). We have also enucleated the eyes of two normal rats weighing 100 gm., but with no effect upon the testicular tissue of the same. To the above rats another 12 animals should be added which received grafts from rats 18–22 days old and which are still alive (see Table 2).

Four to nine whole anterior pituitaries from males 13–22 days of age were grafted at different time-intervals in the anterior eye chamber and, as the necessity arose, under the kidney capsule. If the host animal responded to the first batch of grafts (usually three or four whole anterior pituitaries) no further grafts were given to it. Out of 38 grafted rats, 13 or 34 per cent either had their retracted testes returned into the scrotal sac or when freshly hypophysectomized animals were grafted, incipient involution was arrested, thereafter followed by a restoration that was close to normal.

Some of the definitive results so far obtained have been presented in Table 1. In Table 2 are collected the results for the animals that received grafts from rats 18–22 days old and are still alive. To the grafted rats that sired a normal female should be added two more animals which were killed, but belonged to a still different batch of rats. This raises the number of rats siring normal females to 11. As a matter of fact it seldom happens that, once the process of restoration of the testes becomes definitely stabilized, a grafted male fails to sire a female.

For the benefit of those who wish to go deeper into the problem of the functional activity of the pituitary grafts we should recommend the observance of the following points.

(1) *Mass of the grafted tissue.* When three whole anterior pituitaries of infantile rats are grafted into hypophysectomized hosts, the testes of the recipient animal may or may not respond to the grafted pituitary tissue. Some hosts failing to respond to the first batch of grafts will do so if a second batch is given to them. Some of the hosts that failed to react to a second batch may react if a third batch is grafted under the kidney capsule. These observations have led us to believe that there may be a threshold sensitivity on the part of the testicular tissue to the hormones manufactured by the grafted pituitary tissue. A certain percentage of the grafted rat

Table 1. HYPOPHYSECTOMIZED GRAFTED RATS WITH ENUCLEATED EYES

No. of batch	No. of animals	Body-weight (gm.)			Weight of testes (gm.)		Result of mating to normal females	Size of litter	Remarks
		At hypophysectomy	Before removal of grafts	When killed	Left testis before removal of grafts	Right testis after removal of grafts			
V–58/59	1	110	180	150	0·800	0·158	+	6	—
VII–58	1	97	185	143	1·074	0·543	Not mated	—	Animal had to be killed because of illness
IX–58/59	1	100	203	140	1·212	0·155	+	10	—
IX–58/59	1	97	170	141	0 954	0·182	+	9	—

Table 2. HYPOPHYSECTOMIZED GRAFTED RATS STILL UNDER EXPERIMENT WITH GRAFTS OBTAINED FROM MALES 18–22 DAYS OLD

No. of batch	No. of animals	Average body wt. at hypophysect. (gm.)	Average body wt. Oct. 20, 1959 (gm.)	Condition of testes			Result of mating		Remarks
				Good	Can be palpated	Retracted	Mated	Sired normal females	
X–58/59	5	97	234	4	—	1	4	4	—
XI–59	2	102	242	2	—	—	1	1	—
XII–59	3	99	173	1	2 recovering	--	—	—	These animals belong to a batch recently subjected to experiment
XIII–58/59	2	102	235	1	—	1	1	1	First grafted two months following hypophysectomy

testes showed no sign of stimulation even after three or more consecutive graftings. In these animals, however, the adrenal glands very often grew three to four times the size of the same glands in the hypophysectomized controls. We are inclined to believe that, in so far as our strain of rats is concerned, individual animals may show different threshold sensitivities to gonadotropic hormones, detectable only in situations where the hormone supply is very low.

(2) *Age of the donor of the grafts.* The most satisfactory results were obtained when the grafted tissue was taken from rats 19–22 days old. Pituitaries taken from animals 13–17 days of age gave good grafts. Such grafts stimulated the growth of the host animal very markedly, but their action on the male reproductive organs was less marked than when pituitaries of slightly older animals were transplanted into hypophysectomized hosts. This difference in functional activity between the younger and the older pituitary grafts may probably be explained by a difference in their functional capacity at transplantation in consequence of a difference in the degree of their differentiation.

Pituitaries taken from adult rats (50–60 days old) gave poor results because the grafts did not 'take' well. It is known that tissues of older animals give poorer grafts. Our experimental animals belonged to a random inbred strain, and a certain number of grafted pituitaries, both infantile and adult, underwent early degeneration. In order to avoid reactions attributable to immunity phenomena, it is highly desirable that all the experimental animals belong to a closely inbred strain.

(3) *Time of grafting following the hypophysectomy.* If grafting of the pituitary tissue immediately follows hypophysectomy a greater percentage of the grafted animals will have their reproductive activities restored. Some of the grafted rats, however, had their reproductive capacity restored when grafting of the pituitary tissue took place two months following hypophysectomy. This fact seems to show that prolonged hypophysectomized states may render the repairing action of the pituitary grafts on the reproductive organs more difficult.

(4) *Health of the grafted tissue.* If the pituitary tissue following grafting retains its original size, that is, if it is well supplied with blood, a large percentage of the grafted rats will have their testes partially or almost completely restored. Very few hosts with poor pituitary grafts will show an activation of the testes. Some of the grafted pituitaries may retain their healthy appearance for long periods, and then undergo a very slow degeneration. This process may sometimes go on for many months. Curiously enough, the deterioration of the grafts does not run parallel with a second involution of the testes.

(5) *Health of the grafted animal.* The health of the animal under experiment cannot be sufficiently emphasized. The pituitary grafts will never restore the body functions of the experimental animal to its normal state. In spite of that, some of the grafted rats could not be told apart from normal controls unless their previous life-history had been known. However, when the grafted tissue is removed from such animals they invariably return to their original hypophysectomized state. Animals with seriously affected lungs are practically worthless and should be rejected. Long experience has shown us the importance of the role taken by the operational procedures (anæsthesia, techniques of hypophysectomy and grafting, etc.) one is obliged to use in this experiment, and that the degree of precision of each of them may account for all the difference between failure and success.

A paper dealing *in extenso* with technical procedures and with the action of the pituitary grafts upon their end-organs will be published elsewhere.

[1] Martinovitch, P. N., *Bull. Acad. Serbe des Sciences IV, Sci. Nat.*, 2 (1952).

[2] Courrier, R., *Arch. de Biol.*, 67, 461 (1956). Courrier, R., and Colonge, Andrée, *C.R. Acad. Sci., Paris*, 245, 388 (1957).

[3] Fry, E., and Long, C. N. H., Résumés des communications, Twentieth Cong. Int. Physiol., Bruxelles, 307 (1956).

[4] Harris, G. W., Monographs Physiol. Society, No. 3 (1955).

[5] Benoit, J., and Assenmacher, I., *J. Physiol.*, 47, 427 (1954).

LETTERS TO THE EDITORS

PHYSICS

Enhancement of the High-Spacing Meridional Reflexions in the X-ray Photograph of Keratin impregnated with Heavy-Metal Salts

ENHANCEMENT of the reflexions corresponding to certain orders of the fundamental longitudinal periodicity of 198 A. in the X-ray diffraction photograph of keratin treated with heavy-metal salts has been reported by Fraser and Macrae[1]. They showed, for example, that mercury-treated Lincoln wool fibres gave enhanced seventh, twelfth, sixteenth and nineteenth orders, but they failed to observe any such effect in fibres treated with silver nitrate.

In connexion with parallel electron microscope work being done in this laboratory[2], we have recently performed experiments in which various keratin fibres (Lincoln wool, mohair, human hair and pig bristle) have been impregnated with mercury acetate or silver nitrate after they had been treated with thioglycollic acid to reduce the disulphide groups. With the mercury, we find considerable differences between the diffraction effects produced in the different fibres, and would add the third, fourth and sixth orders to the list of those which can sometimes be enhanced.

Our most significant observations, however, have been made on the Lincoln wool fibres containing silver, for in these we find a quite exceptional enhancement of all the orders from the third to the tenth. Assessment of the absolute intensity of the scattering is made difficult by the high absorption of the metal, which makes a considerable reduction in specimen size necessary if X-rays are to be transmitted with reasonable intensity. However, if the specimen size and exposure are adjusted so that the conventional large-angle photograph can be discerned, the intensity of the scattering at low angles (corresponding to Bragg spacings greater than 20 A.) is so great that details of the discrete reflexions tend to be obscured; when the exposure is adjusted to allow these details to be observed, the photographic film is completely blank at higher angles of scattering.

To a first approximation, therefore, we can say that the effect of the silver is to enhance the meridional reflexions for orders up to the tenth at the expense of the higher orders. This can only mean that the silver is spread out at fairly wide intervals along the fibre axis. When the polarization and Lorentz factors are taken into account, it turns out that the eighth-order reflexion is the strongest by a factor of 2·5 at least; the tenth, sixth and third orders are also strong, and the fourth, fifth, seventh and ninth are present but relatively weak. The absence of the higher orders and the very great intensity of the eighth can be interpreted to mean that the silver atoms are distributed with some regularity at intervals of approximately 25 A. along the α-helix.

Whether the silver is taken up within or on the surface of the microfibril is at present being investigated with the electron microscope, and preliminary results (Sikorski, J., unpublished work) suggest that, as occurs with osmium staining, the metal is for the most part located on and between the microfibrils rather than in them. Since the diffraction effect described is conditional upon the reduction of the disulphide groups, it is tempting to suppose that in some parts of the fibre, at least, the cystine residues occur at intervals of 25 A. along the axis; it may be relevant that a relatively strong eighth-order reflexion occurs in untreated human hair[3] and Lincoln wool[4], but only a weak one in porcupine quill[3], which has a much lower sulphur content.

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[1] Fraser, R. D. B., and Macrae, T. P., *Nature*, 179, 782 (1957).

[2] Sikorski, J., and Simpson, W. S., *Nature*, 182, 1235 (1958).

[3] Bear, R. S., and Rugo, H. J., *Ann. N.Y. Acad. Sci.*, 53, 461 (1951).

[4] MacArthur, I., *Nature*, 152, 38 (1943).

Meridional Reflexions in the X-ray Diffraction Photographs of α-Keratin

X-RAY diffraction photographs of various keratins taken with fine-slit collimators of high resolution display a rich series of meridional reflexions[1-3]. Their spacings may conveniently be regarded as orders of a macro-period of 198 A. with very few exceptions. Astbury[4] has used the spacings and intensities of reflexions reported by Bear and others, from porcupine quill tip, to derive a one-dimensional Patterson diagram, and to explore the possibility of finding the sequence of amino-acid residues in the protein chains of the crystalline portion of the keratin. It might well be that the differences among keratins could provide useful clues in this quest, and the present intention is to direct attention to them.

Photographs were taken using copper $K\alpha$ radiation in an evacuated camera at specimen-film distances of

Table 1. RELATIVE INTENSITIES OF MERIDIAN AND NEAR MERIDIAN REFLEXIONS

Order of 198 A.	Bear, Astbury	Group *A*	Group *B*	Group *C*
3	6	6	6	6
4	1	2	1	2
5	2	1 (38 A.)	2 (38)	0
6	0	0	2	0
7	4	5 (27 0)	5	3
8	2	3	5	6
9	2	1	1	1
10	4	5	5	4
11	3	5	0 or 1	0 or 1
12	0	1	1	0 or 1
13	1	2	2	2
14	0	1 (13·9)	0	0
15	1	3	1	1
16	4	5	3	3*
17	0	0	0	0
18	0	2	0	1
19	3	2	2	2

* Pig bristle 4, human hair 2, horse hair 3.

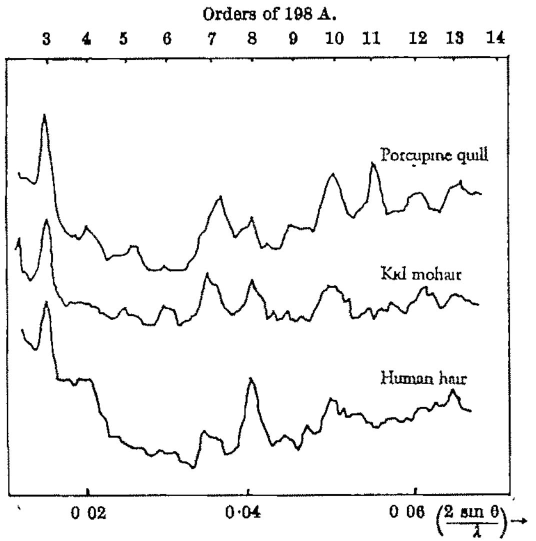

Fig. 1

24 and 20 cm. The six forms of α-keratin studied gave photographs that fall into three groups: group *A*, porcupine quill tip; group *B*, Lincoln wool and kid mohair; group *C*, human hair, horse hair and pig bristle. The intensities of the reflexions observed are reported in Table 1, using Bear's scale of intensities. In exceptional cases the spacing is given in brackets where the reflexion is displaced from the position appropriate to an order of 198 A.

It will be noted that the intensities for group *A* (porcupine quill tip) do not differ greatly from those reported by Bear and used by Astbury; the intensities for group *C* show differences in detail from those of Bear and Rugo, although the general trend is similar. The chief differences among the present groups are in orders 5, 6, 7, 8, 10, 11, 15 and 16. The intensities, similarities and differences observed by eye are confirmed by photometer traces, of which Fig. 1 shows typical examples. Further work is proceeding to investigate the possibility of extending the tentative classification used here to other varieties of keratin fibres.

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[1] MacArthur, I., *Nature*, 152, 38 (1943).

[2] Bear, R. S., *J. Amer. Chem. Soc.*, 66, 2043 (1944).

[3] Bear, R. S., and Rugo, H. J., *Ann. N. Y. Acad Sci.*, 53, 461 (1951).

[4] Astbury, W. T., Proc. Int. Wool Text. Res. Conf. Australia, *B*, 202 (1955).

Thermo-magnetic Effects in Liquids

Recently, several authors[1-7] have investigated the effect of a magnetic field on thermal convection in electrically conducting liquids. In this communication we present the results of a simple experiment on the effect of the magnetic field on the rate of cooling of two liquids, namely, mercury and water. A glass tube (length 20 cm. and diameter 4·2 cm.) was placed on a frame suspended between the pole-pieces of a large electromagnet (type *ADL* Cambridge, Mass, U.S.A.). The frame was made of wood and brass rods, and was completely isolated (thermally) from the pole pieces of the magnet. The glass tube, filled with mercury up to a height of 15·7 cm., was heated and then allowed to cool in a magnetic field of about 23,000 oersteds and without the magnetic field. The cooling curves for both cases are shown in Fig. 1. These curves clearly show that the rate of cooling for mercury is faster in the magnetic field. The experiment was later repeated also with water, and a similar effect on the rate of cooling was observed (Fig. 1).

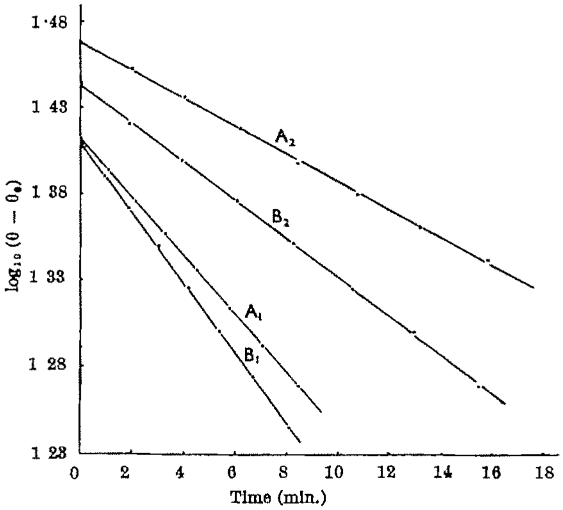

Fig. 1. Cooling curves for mercury and water with and without the magnetic field. Curves A_1 and B_1 refer to mercury, while A_2 and B_2 are for water. Curve *B* corresponds to the cooling in a magnetic field of ~23,000 oersteds and *A* refers to field free cooling. Abscissa denotes time in minutes and ordinate $\log_{10}(\theta - \theta_0)$, where θ is the instantaneous temperature of the liquid and θ_0 is that of the surroundings

In another experiment with water as the liquid, the rate of cooling was investigated in the presence of the magnetic field, which was switched off for a few minutes and later re-established. The slopes of the cooling curves obtained in the magnetic field are essentially the same, and are greater than that of the curve obtained while the magnetic field was switched off. This further supports the finding that the cooling becomes faster in the presence of a high magnetic field. Experiments were also performed to study the effect of a magnetic field on the rate of heating for both the liquids, and it was found that the rate of heating is also faster in the presence of a magnetic field. Similar results were also obtained when the glass was replaced by a copper container.

From these experiments we conclude that the rate of cooling and also the rate of heating for these liquids increases in the presence of a high magnetic field.

The above results may possibly be explained on the basis of the change in specific heat of the liquid in a magnetic field or they may be due to a change in its absorptivity (or emissivity) in the presence of a magnetic field. In the case of mercury, however, there should be some additional effect due to the inhibition of convection in the magnetic field.

The nature of the variation of the rate of cooling of liquids subjected to different magnetic fields will be the subject of future investigation.

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[1] Lehnert, B., *Proc. Roy. Soc.*, A, **233**, 299 (1955).
[2] Nakagawa, Y., *Nature*, **175**, 417 (1955).
[3] Nakagawa, Y., and Frenzen, P., *Tellus*, **7**, 1 (1955).
[4] Fultz, P., and Nakagawa, Y., *Proc. Roy. Soc.*, A, **231**, 211 (1955).
[5] Jirlow, K., *Tellus*, **8**, 252 (1956).
[6] Levengood, W. C., *Nature*, **177**, 631 (1956).
[7] Nakagawa, Y., *Proc. Roy. Soc.*, A, **240**, 108 (1957).

CHEMISTRY

Effect of Ultrasonic Insonation on the Carbon Tetrachloride–Water–Dissolved Helium System

IT was the purpose of this study to obtain more evidence regarding the effect of ultrasonics on chemical reactions, and to determine the relationship between the reaction yield and solubility of dissolved gas in the system. Griffing[1] found that gases of different solubilities have different thresholds of cavitation; but no attempt was made to ascertain the effect of varying the solubility of a particular gas.

The system studied was the carbon tetrachloride–water–dissolved helium system. In order to ensure that the helium was the gas responsible for any reaction that might be observed, the water used was purified and degassed. Carbon tetrachloride was added and the mixture was vigorously agitated. Helium was then bubbled through, and after settling the substrate consisted of two distinct phases. The phase rich in carbon tetrachloride was used for the experiments. The carbon tetrachloride–water–dissolved helium mixture was then transferred (in a helium atmosphere) to a specially designed reaction vessel, one side of which was covered with a polyester film of high sound permeability. This served as a sound window.

The reaction mixture was exposed to 1,000 kc./s. sound energy at a power output of 7·20 watts/sq. cm. The sound was transmitted through a plain tap water medium from crystal to vessel. This water was circulated and served also as a constant-temperature bath. The distance between the crystal and reaction vessel surface was maintained the same for each run. This assured uniform sound absorbance by the sample. The samples after insonation were analysed for chlorine by addition of potassium iodide solution and the titration of the liberated iodine with sodium thiosulphate solution. Starch was used as indicator. It was found that the *p*H change brought about by the reaction was sufficient to supply the acid medium need for the above analysis.

Samples which were insonated immediately after they were transferred from the preparation apparatus to the reaction vessel gave erratic results. Samples which were transferred and allowed to stand 12 hr. before insonation gave much greater reproducibility. Griffing[1] observed a similar phenomenon. Different final concentrations of chlorine, due to insonation, were observed for vessels which were completely filled and those which were partially filled with reaction mixture (the vapour space being filled with helium). In addition to chlorine, it was thought that trace amounts of hexachloroethane could be detected.

The formation of chlorine and hexachloroethane from the reaction mixture, in the absence of oxygen, indicates a free-radical mechanism. This could well be due to the break-up of the water molecule as a result of local heating due to the adiabatic compression of the dissolved gas, as Griffing has suggested[2].

The different yields observed for partially filled and completely filled vessels can be ascribed to the variation in gas solubility with pressure. Ultrasonic insonation is a degassing process. It is obvious that the residual pressure in a vessel which was originally filled is much greater than the pressure in a vessel which was partially filled. According to Griffing's theory with regard to the effect of solubility on cavitation[1], the yield of chlorine would be greater at low pressures and lower at high pressures. The data show that the yield for filled vessels was lower than that for partially filled vessels. This supports the solubility theory of Griffing, using only a single gas. This eliminates any doubt which might have arisen due to the varying of solubility with different gases and thereby changing other physical properties as well.

The cause of the erratic results after sample transfer was probably nucleation—the formation of centres of gas bubble agglomeration. This would occur in random fashion for each run, thereby introducing a lack of similarity in sample preparation.

I would like to acknowledge the help of Dr. O. L. Updike during the course of the work, which was carried out at the University of Virginia, Charlottesville, Virginia.

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[1] Griffing, Sette, Sullivan and Majowicz, "Experiments on Cavitation and Related Reactions" (Catholic University, Washington, D.C.) (unpublished).
[2] Griffing and Fitzgerald, *J. Chem. Phys.*, **25**, 5 (1956).

Optical Rotatory Dispersion Studies on Partially Methylated Cellulose

ROTATORY dispersion studies have been conducted on a large number of proteins and polypeptides[1–3]. We wish to report some preliminary studies on the rotatory dispersion of polysaccharides. The material used for this work was 'Methocel' (registered trade mark of the Dow Chemical Company, Midland, Michigan) supplied by the Dow Chemical Co. 'Methocel' is a partially methylated cellulose which has the characteristic of being more soluble in cold water than in hot water. This investigation was carried out on material with an intrinsic viscosity of 1·6 (corresponding roughly to a molecular weight of 35,000) and a methyoxyl analysis of 29 5 per cent (1·8 OCH_3 groups/anhydroglucose unit). The method of solubilizing this particular polysaccharide consisted in dispersing the material in boiling water and allowing it to cool to room tem-

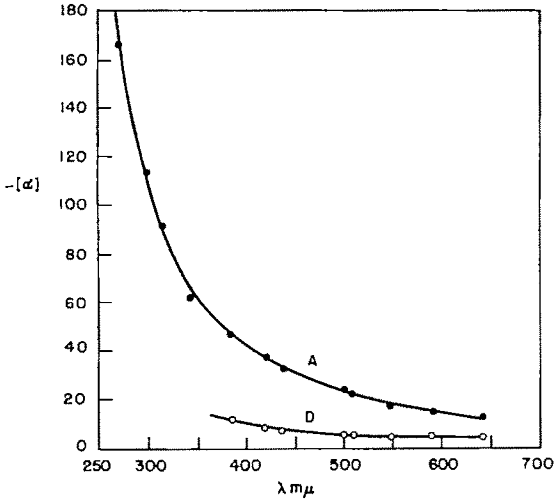

Fig. 1. Optical rotatory dispersion curves of 2 per cent aqueous solutions of methylcellulose at 24° C. See text for description of curves *A* and *D*

perature. Solution *A* (Figs. 1 and 2) was further cooled to 0° C. overnight and then maintained at room temperature (24° C.). Solution *D* (Figs. 1 and 2) was maintained at room temperature. The dispersion curves for these two solutions are shown in Fig. 1. The majority of the measurements were carried out on a Keston photoelectric polarimeter attached to the Beckman *DU* spectrophotometer. The cell was covered with black paper to eliminate any spurious rotation as described in a recent paper by Winkler and Markus[4]. The dispersion curve of solution *A* from 270 mμ to 600 mμ was obtained on a Rudolph photoelectric spectropolarimeter, model 200 *S*. (The use of this instrument was kindly permitted by Prof. M. L. Wolfrom of the Chemistry Department, Ohio State University, Columbus, Ohio.) The results using the Keston instrument checked very well with the results on the Rudolph instrument.

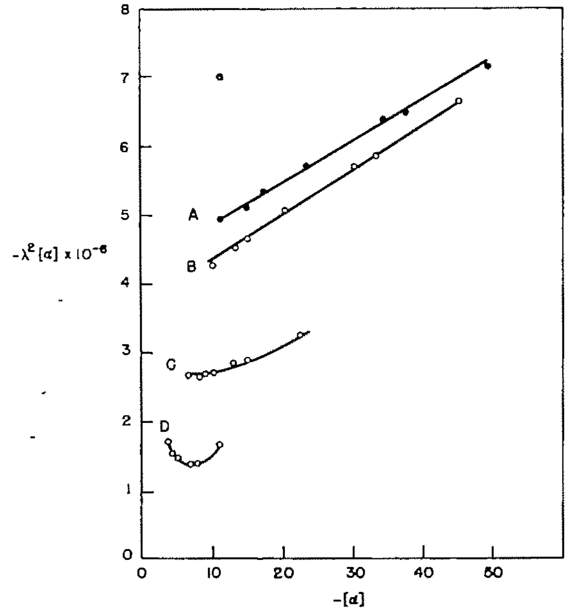

Fig. 2. Modified Drude plots of the dispersion data in Fig. 1. See text for description of curves *A*, *B*, *C* and *D*

The dispersion data were plotted by the method of Yang and Doty[2] as given in the following equation:

$$\lambda^2[\alpha] = \lambda_c^2[\alpha] + K \qquad (1)$$

(λ denotes wave-length in mμ, [α] specific rotation, λ_c the dispersion constant and *K* the intercept).

The modified Drude plots of the dispersion data are given in Fig. 2. The partially methylated cellulose solubilized at 0° follows the simple one-term Drude equation as shown by curve *A*, Fig. 2. Curve *D*, Fig. 2, is the complex dispersion of this particular polysaccharide which had been solubilized at room temperature. This solution is very unstable and progresses from curve *D* to curve *C* (1 hr. in the polarimeter tube) to curve *B* (16 hr. in the polarimeter tube). Curve *B* represents an equilibrium state. This equilibrium solution may be shaken and the original curve *D* is returned. This anomalous dispersion can be explained on the basis of molecular association causing an aggregate which slowly settles out of solution leaving the randomly dispersed material behind. By analogy with the polypeptide studies of Yang and Doty[2] the linear Drude plots (curves *A* and *B*, Fig. 2) represent partially methylated cellulose randomly dispersed in solution.

These studies will be further amplified in a detailed publication on this phenomenon.

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[1] Doty, P., and Yang, J. T., *J. Amer. Chem. Soc.*, 78, 498 (1956).

[2] Yang, J. T., and Doty, P., *J. Amer. Chem. Soc.*, 79, 761 (1957).

[3] Jirgensons, B., *Arch. Biochem. Biophys.*, **74**, 57, 70 (1958); 78, 227, 235 (1958).

[4] Winkler, M. H., and Markus, G., *J. Amer. Chem. Soc.*, 81, 1873 (1959).

Deuterium–Hydrogen Exchange between Water and Macromolecules: Accessibility of Cellulose

Although many workers have observed the exchange reaction of heavy water with macromolecules indirectly by the change in composition of the water[1,2], and by infra-red measurements[3-5], only one group has reported the measurement of the exchange directly by the change in weight of the macromolecules. Burley, Nicholls and Speakman[6] measured the increase in weight when wool keratin reacted with the vapour of heavy water. They used a quartz spring balance with a small sample (about 0·1 gm.). In the present work, in which the gravimetric method was also employed, a much larger sample (about 6 gm.) was used. The sample was placed in a glass cell similar to one used in the measurement of adsorption isotherms of water on fibrous macromolecules[7].

The principal problem in the gravimetric method is to achieve complete dryness. Recently, the conditions necessary for obtaining this dryness were determined unequivocally by Lang and Mason[8] for cellulose and by Hvidt and Linderstrøm-Lang[9] for insulin. Complete drying was obtained in the present work by a combination of diffusion and rotary vacuum pumps and 'dry ice'-acetone baths. The celluloses (cotton and cotton mercerized in 18 per cent sodium hydroxide) were evacuated for a minimum of 72 hr. at 70° C., although there was usually no change in weight after 24 hr.

Table 1

Cellulose	Weight increase of deuteration (per cent)	Heat of wetting (refs. 7 and 9) (cal / gm.)	Monolayer values (refs. 7 and 9) (gm. water/100 gm.)		Accessibility (ref. 10)
			Brunauer–Emmett–Teller	Harkins–Jura	
Cotton	0·710	12·33	3·19	3·29	0·38
Mercerized cotton	1·21	20·25	5·21	5·36	0·65
Ratio, M/C	1·71	1·64	1·63	1·63	

Deuterium loading was obtained by keeping the celluloses in contact with heavy-water vapour (99·7 per cent) for several hours, and then evacuating to constant weight. After four to six loadings, there was no further increase in the evacuated dried weights. The process was completely reversed by ordinary water vapour, so that there were no 'resistant' deuteroxyl groups produced[3]. The percentage gains in weight along with other results for the same substances[7,9] are given in Table 1.

Included in Table 1 are the accessibilities of the two celluloses. The accessibility is defined[10] as the ratio of the number of hydroxyl hydrogens exchanged to the total number in the cellulose. The accessibility of cotton cellulose to the vapour of heavy water is exactly the same as that calculated by Valentine[11] for the amorphous fraction. Valentine based his calculations on correlations between infra-red measurements of celluloses deuterated with heavy-water vapour[3] and water vapour adsorption isotherms[10]. The present accessibility of cotton cellulose is also similar to that for bacterial cellulose[3].

The ratio of the Brunauer–Emmett–Teller monolayer molar values[7,9] to the moles of deuterated hydroxyls are 0·25 and 0·24 for cotton and mercerized cotton respectively. This suggests that each accessible glucose anhydride unit adsorbs about one molecule of water in the complete monolayer.

Of the three hydroxyls in the glucose unit, two are on the ring and one is on the side-chain. The tentative deduction that a maximum of one molecule is adsorbed directly on each accessible glucose unit suggests in turn that the side-chain hydroxyls may be the sites of hydrogen bonding[7], and that they may be exchanged more rapidly than the ring hydroxyls. Kinetic experiments in progress appear to support the latter hypothesis.

This work was reported at the annual conference of the Chemical Institute of Canada, May 25–27, 1959, Halifax, N.S.

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[1] Frilette, V. J., Hanle, J., and Mark, H., *J. Amer. Chem. Soc.*, 70, 1107 (1948).

[2] Hvidt, A., and Linderstrem-Lang, K., *Biochim. Biophys. Acta*, 16, 168 (1955); *C.R. Lab. Carlsberg*, Ser. Chim., 29, 385 (1955).

[3] Mann, J., and Marrinan, H., *J. Trans. Farad. Soc.*, 52, 481, 487, 492 (1956).

[4] Almin, K. E., *Svensk Papp.-Tidn.*, 55, 767 (1952).

[5] Fraser, R. D. B., and MacRae, T. P, *J. Chem. Phys.*, 28, 1120 (1958).

[6] Burley, R. W., Nicholls, C. H., and Speakman, J. B., *J. Text. Inst.*, 46, *T*427 (1955).

[7] Morrison, John L., and Dzieciuch, M. A., *Canad. J. Chem.*, 37, 1379 (1959).

[8] Lang, A. R. G., and Mason, S. G., *Canad. J. Chem.*, 37, 1829 (1959).

[9] Morrison, John L., and Chapman, R. (unpublished results).

[10] Howsmon, J. L., *Text. Res. J.*, 19, 152 (1949).

[11] Valentine, L., *J. Polymer Sci.*, 27, 313 (1958).

A Proposal for the Nomenclature of Free Radicals

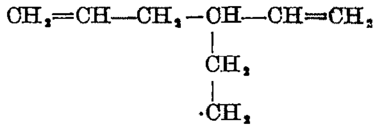
IN the course of preparation of a number of organic free radicals for mass spectrometric and electron spin resonance studies, it has become apparent that the system of nomenclature for organic compounds approved by the International Union of Chemistry[1] does not, in many cases, serve to identify a free radical unambiguously. As an example, there are listed below the possible butenyl radicals, the dot indicating the nominal position of the free electron. (This is not to imply that the electron is actually localized at this site.)

(1)	$CH_3CH=CH\dot{C}H_2$	2-butenyl
(2)	$CH_3CH=\dot{C}CH_3$	
(3)	$CH_2=CHCH_2\dot{C}H_2$	1-butenyl
(4)	$CH_2=CH\dot{C}HCH_3$	
(5)	$CH_2=\dot{C}CH_2CH_3$	
(6)	$CH=CHCH_2CH_3$	
(7)	$CH_2=C(CH_3)\dot{C}H_2$	2-methyl propenyl
(8)	$\dot{C}H=C(CH_3)CH_3$	

The difficulty does not arise with derivatives of the radicals; for example, the chloride of (4) is 3-chloro 1-butene, and that of (8) is 1-chloro 2-methyl propene. When, however, the corresponding 'free' radicals are considered the system fails for lack of a functional group name for the free electron. We propose that the ending 'yl' be retained for the free radical and that the free electron be given the functional group name of 'el', preceded by a number to indicate its position as for any other substituent. Thus the free radical (4) becomes 1-butenyl 3-el as distinguished from (5) 1-butenyl 2-el.

The naming of di-radicals would follow the usual practice for double bonds; for example, $\dot{C}H_2-CH=\dot{C}H$ would be called propenyl 1,3 di-el and $\dot{C}H=C=\dot{C}H$ propadienyl 1,3 di-el.

A free radical in which the free electron is located on a side-chain, such as the (hypothetical) radical



$$CH_2=CH-CH_2-\underset{\underset{\cdot CH_2}{|}}{\underset{CH_2}{\underset{|}{C}H}}-CH=CH_2$$

would be named in this system (4 ethyl 2-el) 1,5 hexadiene.

At present radicals such as (1), (4) and (7) can be named as derivatives of allyl; (1) is γ-methylallyl, (4) is α-methylallyl and (7) is β-methylallyl. These names, although useful in indicating the relation of these radicals to each other as derivatives of allyl by methyl substitution, may cause confusion since derivatives of the butenes are sometimes named from their relation to α-butylene and β-butylene, in which the prefix refers to the position of the double bond rather than that of substitution.

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[1] *J. Amer. Chem. Soc.*, 55, 3905 (1933).

BIOCHEMISTRY

Enzymatic Cycle of Inorganic Nitrogen in Animal Tissues

It has previously been demonstrated that oximino compounds are widely distributed in organisms[1], and that two enzymes, named oximase[2] and transoximase[3], which participate in oxime metabolism, exist in some animals. In pursuing the behaviour of hydroxylamine in living cells, we have now established that there is an enzymatic cycle between ammonia and nitrate.

An example of the procedure of preparing the enzyme solution was as follows: 30 gm. of hen liver were homogenized with 90 ml. of 0·1 M phosphate buffer of pH 7·5, and after 30 min. at 35° C., the homogenate was centrifuged at 3,000 r.p.m. for 10 min. at 0° C. The supernatant was treated with acetone at −10° C., and the precipitate which was produced at 33–55 per cent concentration of acetone was gathered by centrifuging at 4,000 r.p.m. for 7 min. 1 gm. of this precipitate was dissolved in 20 ml. of water, dialysed against running water for 3 hr. and centrifuged at 3,000 r.p.m. for 7 min. The supernatant was used as enzyme solution; it contains about 0·2 mgm. protein nitrogen in 1 ml. As the enzyme is labile, the solution must be prepared daily from fresh liver.

To measure the reducing action of enzyme preparation, 1 ml. of 0·001 M substrate was mixed with 1 ml. of enzyme solution, 1 ml of 0·013 per cent reduced diphosphopyridine nucleotide solution, 0·5 ml. of 0·005 per cent flavin adenine dinucleotide solution and 0·5 ml. of 0·1 M phosphate buffer of pH 7·5. A mixture containing no substrate served as control. The reaction temperature was 30° C. and the time 20 min. The enzymatic activity was determined spectrophotometrically and expressed in decrease (−) of absorption (−log T) at 340 mμ. Some of the results obtained are given in Table 1.

Nitrate, nitrite, hyponitrite and hydroxylamine are thus reduced by an enzyme system from hen liver. It was further found that the enzymes manifest the highest activity at pH 7·7 and 30° C., and that they are completely destroyed by heating at 50° C. for 5 min. Reduced triphosphopyridine nucleotide and flavin mononucleotide do not act as coenzymes for the reduction by liver enzymes.

Recently, hyponitrite reductase was demonstrated in *Neurospora* by Medina and Nicholas[4], and hydroxylamine reductase was extracted from a halotolerant *Micrococcus* by Taniguchi, Sato and Egami[5] as well as from *Neurospora* by Zucker and Nason[6]. It remains to be seen whether the enzyme system of hen liver contains the same reductases as those found in microbes.

For estimating the dehydrogenating action of liver enzyme system, the freshly prepared enzyme solution was heated beforehand at 40° C. for 5 min., because the reductases are more easily inactivated by heating than the dehydrogenases. The reaction mixture consisted of 1 ml. of 0·001 M substrate, 1 ml. of enzyme solution, 1 ml. of 0·013 per cent diphosphopyridine nucleotide solution and 1 ml. of 0·05 M *tris*-buffer of pH 7·5. The control contained no substrate. The increase (+) of −log T at 340 mμ was determined after a 20 min. incubation at 30° C. Some of the results are given in Table 2.

Table 1 Reducing Action of Liver Enzyme System

Substrate:	$NaNO_3$	$NaNO_2$	$Na_2N_2O_2$	NH_2OH	$(NH_4)_2SO_4$	Without substrate
−log T	−0·105	−0·148	−0·124	−0·122	−0·024	−0·022

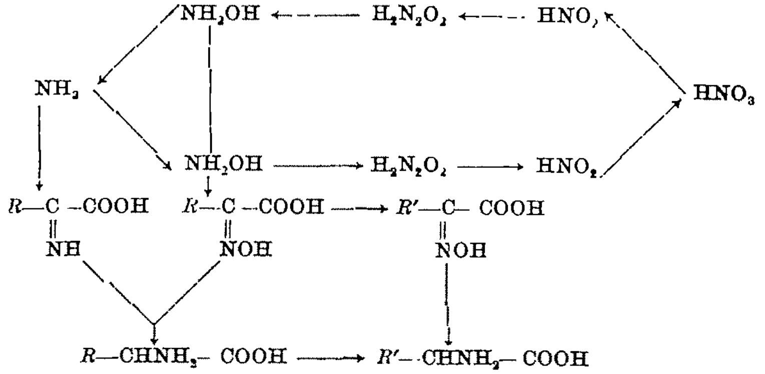
Table 2. Dehydrogenating Action of Liver Enzyme System

Substrate:	$(NH_4)_2SO_4$	NH_2OH	$Na_2N_2O_2$	$NaNO_2$	$NaNO_3$	Without substrate
−log T	+0·120	+0·095	+0·138	+0·084	+0·021	+0·015

Ammonia, hydroxylamine, hyponitrite and nitrite are thus dehydrogenated by the liver enzyme preparation. The dehydrogenation, however, does not proceed in a mixture containing phosphate buffer. It was further observed that the enzymes display the highest activity at pH 7·3–7·5 and 30° C., and that they are destroyed by heating at 60° C. for 5 min. Triphosphopyridine nucleotide, flavin adenine dinucleotide and flavin mononucleotide show no coenzymatic action for the dehydrogenation.

Klausmeier and Bard[7] reported recently that an ammonium dehydrogenase is present in *Bacillus subtilis*. However, both tri- and di-phosphopyridine nucleotide act as coenzymes for the bacterial dehydrogenase and ammonium sulphate does not serve as its substrate.

The enzyme solution was also prepared from silkworms, and its reducing and dehydrogenating actions were studied in the same way. Similar results were obtained.



Scheme 1. Inorganic nitrogen cycle and amino-acid formation

The combination of inorganic nitrogen cycle and amino-acid formation, including the transoximase–oximase reaction, can thus be formulated as in Scheme 1.

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[1] Yamafuji, K., Kondo, H., and Omura, H., *Enzymologia*, 14, 153 (1950).

[2] Yamafuji, K., *Nature*, 167, 770 (1950). Yamafuji, K., Aoki, M., and Omura, H., *Enzymologia*, 17, 371 (1956)

[3] Yamafuji, K., *Nature*, 171, 745 (1953). Yamafuji, K., and Eto, M., *Enzymologia*, 16, 247 (1953)

[4] Medina, A., and Nicholas, D. J. D., *Nature*, 179, 533 (1957).

[5] Taniguchi, S., Sato, R., and Egami, F., "Symposium on Inorganic Nitrogen Metabolism", edit. by McElroy, W. D., and Glass, B., 87 (Johns Hopkins Press, 1956).

[6] Zucker, M., and Nason, A., *J. Biol. Chem.*, 213, 463 (1955).

[7] Klausmeier, R. E., and Bard, R. C., *J. Bacteriol.*, 68, 129 (1954)

Cysteine-S-sulphonate as an Intermediate in Microbial Synthesis of Cysteine

THE synthesis of cysteine in micro-organisms from inorganic sulphur sources has been studied for some time, and the available evidence suggests that sulphate sulphur is reduced to the thiosulphate or sulphide state prior to its introduction into the carbon chain of cysteine[1-3], although some investigators have postulated that the formation of sulphur–carbon linkage occurs at the sulphite stage[4]. Working with mutant strains of *Aspergillus nidulans*, Hockenhull[2] suggested that thiosulphate is the form in which sulphate sulphur combines with the carbon skeleton. Based on the observations that cysteine-S-sulphonate supported good growth of all the thiosulphate-requiring mutants tested and that growth of these strains on thiosulphate was enhanced by the addition of serine, he concluded that thiosulphate first reacts with serine to form cysteine-S-sulphonate as an intermediate and cysteine is formed from the latter by hydrolysis. The same possibility was also postulated by Shepherd[4], although the involvement of thiosulphate in the main route of sulphate assimilation was denied by this investigator. Schlossmann and Lynen[5], on the other hand, reported the occurrence in yeast of a pyridoxal phosphate-dependent enzyme catalysing the condensation of hydrogen sulphide with serine to form cysteine. The present communication describes more convincing evidence in favour of the cysteine-S-sulphonate pathway in *A. nidulans*.

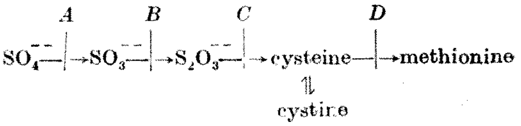
Table 1. GROWTH OF MUTANTS ON VARIOUS SULPHUR SOURCES

Group	No. of mutants	Sulphur source added					
		Na_2SO_4	Na_2SO_3*	$Na_2S_2O_3$*	Cysteine*	Cystine	Methionine
Wild type	—	+	+	+	+	+	+
A	1	—	+	+	+	+	+
B	1	—	—	+	+	+	+
C	6	—	—	—	+	+	+
D	6	—	—	—	—	—	+

+, Growth ; —, no growth (after seven days). The basal medium was supplemented with 100 μgm. per ml. of sulphur compound. All amino-acids used were L-isomers. Incubated at 32° C.

* Sterilized by Seitz filtration.

Mutant strains of *A. nidulans* (I.F.O. Collection No. 5713) requiring methionine for growth were isolated by Fries's filtration technique[6] from conidiospore suspensions exposed to ultra-violet irradiation, and 14 stable mutants were obtained from about 700 isolates. The responses of these strains as well as that of the parent wild-type strain are summarized in Table 1. The basal sulphur-deficient medium used in these experiments contained per litre 30 gm. glucose, 3 gm. sodium nitrate, 1 gm. dipotassium hydrogen phosphate, 0·5 gm. potassium dihydrogen phosphate, 0·2 gm. magnesium chloride, 1 ml. of trace element solution, and 17 gm. of agar (*p*H 6·4). (The trace element solution contained in mgm. per litre: $FeCl_3.6H_2O$, 1,000; $CuCl_2.2H_2O$, 300; $MnCl_2.4H_2O$, 72; $(NH_4)_6Mo_7O_{24}.4H_2O$, 40; $ZnCl_2$, 4,200.) All the growth tests were carried out at least twice. The results obtained indicate that the mutants can be classified into four groups, namely, *A*, *B*, *C* and *D*, according to their nutritional requirements. It appears that the simplest explanation for these findings as well as those reported in previous work[2,3] may be provided by the following scheme of sulphate assimilation:



$$SO_4^{--}\overset{A}{\vert}\rightarrow SO_3^{--}\overset{B}{\vert}\rightarrow S_2O_3^{--}\overset{C}{\vert}\rightarrow \underset{\text{cystine}}{\underset{\upharpoonleft\downharpoonright}{\text{cysteine}}}\overset{D}{\vert}\rightarrow \text{methionine}$$

where the site of block in each group of mutants is indicated by *A*, *B*, etc.

Having assumed the correctness of this pathway, the mechanism of the incorporation of thiosulphate into cysteine was then studied using the 6 strains belonging to the *C* group in which this very reaction is genetically blocked. If this step of assimilation actually involves cysteine-S-sulphonate as an intermediate, it is expected that this step is controlled by at least two genes. This possibility was examined by subjecting the 6 strains to the heterokaryosis test on the basal sulphur-deficient medium supplemented with 200 μgm. of sodium sulphate and 5 μgm. of L-cystine (for heterokaryosis see ref. 7). It was thus found and confirmed by repeated experiments that one strain (designated as 793) can form heterokaryons with any of the other 5 strains (designated as 562, 611, 649, 721 and 831). No signs of heterokaryosis could, however, be detected in any pair of the latter 5 strains, indicating their genetical identity.

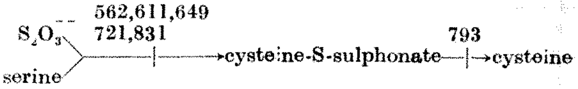
Table 2. UTILIZATION OF CYSTEINE-S-SULPHONATE BY MUTANTS OF THE *C* GROUP

Strain	Wild type	793	562	611	649	721	831
Growth (mgm. dry wt./15 ml.)	57·8	3·0	49·5	37·3	19·0*	39·3	40·8

Average of four experiments. The basal liquid medium was supplemented with 100 μgm. of Seitz-sterilized sodium cysteine-S-sulphonate per ml. Incubated at 33° C. for ten days without shaking.

* This strain showed rather poor growth even in a methionine medium.

Since strain 793 was thus genetically differentiated from the other 5 strains of the *C* group, it seemed of considerable interest to study the ability of these mutants to utilize cysteine-S-sulphonate for growth. The growth experiments were therefore conducted in a liquid medium containing 100 μgm. of sodium cysteine-S-sulphonate (prepared by a modification of the method of Kolthoff and Stricks[8]) per ml. as the sole source of sulphur. The *p*H of the medium was raised to 7·5 to minimize the spontaneous decomposition of the sulphur compound. As can be seen from Table 2, it was clearly shown that strain 793 is quite unable to grow in this medium, whereas the same medium can support good growth of the other 5 strains as well as the wild-type strain. These results, together with those of Table 1, strongly suggest that strain 793 has a genetical block between cysteine-S-sulphonate and cysteine and the other 5 strains are deficient in the enzyme responsible for the condensation of thiosulphate with serine (or other 3-carbon compounds) to form the sulphonate. This situation will be understandable from the following scheme:



$$\left.\begin{matrix}S_2O_3^{--}\\ \text{serine}\end{matrix}\right\rangle\overset{562,611,649}{\overset{721,831}{\vert}}\longrightarrow \text{cysteine-S-sulphonate}\overset{793}{\vert}\rightarrow \text{cysteine}$$

From the evidence reported in this communication, it seems very likely that cysteine-S-sulphonate is involved in the normal pathway of cysteine biosynthesis in *A. nidulans*. The occurrence of this pathway receives further support from the finding that the sulphonate can be detected polarographically in a reaction mixture containing a cell-free yeast extract, thiosulphate, serine and pyridoxal phosphate[9].

We wish to thank Mr. S. Baek for the synthesis of sodium cysteine-S-sulphonate.

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Osaka. Oct. 19.

[1] Lampen, J. O., Roepke, R. R., and Jones, M. J., *Arch. Biochem.*, **13**, 55 (1947).
[2] Hockenhull, D. J. D., *Biochim. Biophys. Acta*, **3**, 326 (1949).
[3] Horowitz, N. H., "Adv. Genet.", **3**, 33 (1950).
[4] Shepherd, C. J., *J. Gen. Microbiol.*, **15**, 29 (1956).
[5] Schlossmann, K., and Lynen, F., *Biochem. Z.*, **328**, 591 (1957).
[6] Fries, N., *Nature*, **159**, 199 (1947).
[7] Pontecorvo, G., *Cold Spring Harbor Symp. Quant. Biol.*, **11**, 193 (1946).
[8] Kolthoff, I. M., and Stricks, W., *J. Amer. Chem. Soc.*, **73**, 1728 (1951).
[9] Baek, S., and Sato, R. (in preparation).

Shape of the Precipitate in Immunoelectrophoresis

IN immunoelectrophoresis, according to Grabar and Williams[1], diffusion of the electrophoretically separated antigens towards the immune serum added in longitudinal basins parallel to the migration axis after the electrophoretic separation will result in the formation of precipitates in the gel, occupying varying positions between antigen and antibody diffusion centres as well as varying electrophoretic positions.

These precipitates generally appear as curved lines with their convexity towards the antibody diffusion centre, because of the immune serum diffusing in a straight line in a direction perpendicular to the electrophoretic axis, while the antigens diffuse from roughly circular diffusion centres in the agar. The point of the precipitate which is nearest to the antibody basin, the peak of the precipitate, is taken to indicate the average electrophoretic position of the electrophoretically separated component giving rise to this precipitate[2].

In some instances, precipitates are found which deviate from the half-moon-shaped symmetrical precipitates by presenting varying degrees of curvature or possessing different degrees of asymmetry. Also, precipitates may be found which have more than one peak. It has been found that these different shapes of the precipitates are remarkably constant for the same precipitating component, when sera from normal individuals are investigated under standard experimental conditions[3,4].

Grabar[5] demonstrated that the γ-globulin precipitate in normal human sera, which incidentally appears as an extended line rather than a curved half-moon-shaped precipitate, consisted of a "family of proteins", each molecule population with its own specific electrophoretic mobility although serologically identical.

In certain human sera, an extended precipitate is obtained in the α_2-globulin region, which therefore according to Grabar's findings might indicate the presence of two or more molecule populations being serologically identical, although presenting differences in electrophoretic mobilities. Also, at least two molecule populations with discrete electrophoretic positions are present, as indicated, by obtaining a precipitate with two peaks if the separation is prolonged[6].

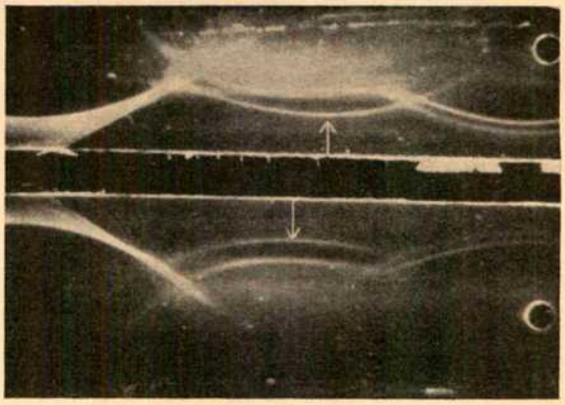

Fig. 1. Immunoelectrophoresis of mixture of sera belonging to *G.c.*1 : 0·1 ml. and *G.c.*2 : 0·4 ml. (upper basin) and mixture of the same sera in equal amounts (lower basin). To the far left the albumin-precipitate is shown and to the right the starting point and the transferrin-precipitate. The group-specific components are indicated by arrows

In other normal human sera either the fast-moving molecule population or the slowly moving molecule population is present. Mixture of sera containing the fast-moving and slowly moving precipitating components in equal amounts will result in the immunoelectrophoretic appearance of a precipitate which is extended or presents two peaks, depending on the separation time and which is indistinguishable from the same precipitate found in certain other normal sera[6]. Also, evidence for the genetic inheritance of these group-specific components has been found[7].

If sera belonging to the fast type (*G.c.*1) and the slow type (*G.c.*2) are mixed in unequal amounts, for example, in the proportions of 1 : 4, an asymmetric precipitate will occur where the peak of the precipitate is displaced from the midpoint towards the cathode (Fig. 1). In a similar way an asymmetric precipitate with its peak displaced towards the anode will be obtained if the fast-moving precipitating component is quantitatively predominant in the mixture.

This seems to demonstrate that the electrophoretic positions of the precipitates can be changed by quantitative factors. This change of electrophoretic position is even more pronounced if sera belonging to different groups are mixed in the proportions 1 : 10. If the slowly moving precipitating compon-

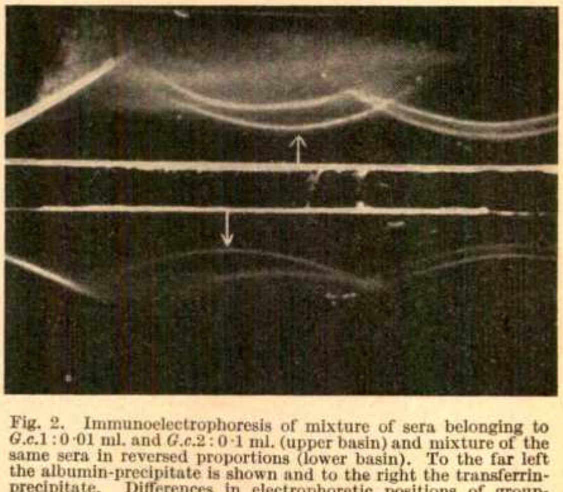

Fig. 2. Immunoelectrophoresis of mixture of sera belonging to *G.c.*1 : 0·01 ml. and *G.c.*2 : 0·1 ml. (upper basin) and mixture of the same sera in reversed proportions (lower basin). To the far left the albumin-precipitate is shown and to the right the transferrin-precipitate. Differences in electrophoretic positions of group-specific components are indicated by arrows

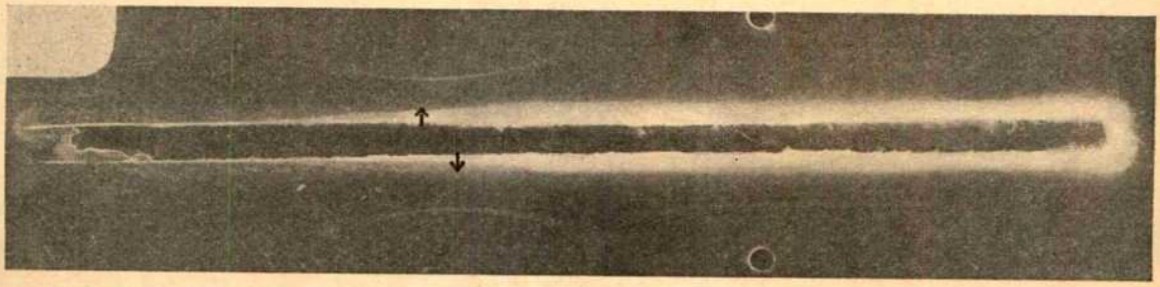

Fig. 3. Immunoelectrophoresis of mixture of sera belonging to *Hp* 1–1 and 2–2 in the same amounts (upper basin) as compared to haptoglobin-precipitate of a serum belonging to *Hp* 2–1 (lower basin). By means of an absorption technique only the haptoglobin-precipitates appear in the α_2-globulin region

ent is quantitatively predominant in the mixture, immunoelectrophoresis will demonstrate a precipitating component occupying an electrophoretic position more towards the cathode than if the fast-moving precipitating component is quantitatively predominant in the mixture (Fig. 2). None the less, the differences are not as pronounced as when the precipitates of unmixed sera belonging to these two different groups are compared.

With the haptoglobins, results of a similar nature have been obtained. These proteins have been demonstrated to present a considerable degree of 'microheterogeneity' in starch-gel electrophoresis. Three main groups can, however, be readily differentiated in starch-gel electrophoresis[8], agar-gel electrophoresis[9] and immunoelectrophoresis[6] although immunologically identical[10]. Mixture of sera belonging to *Hp* 1–1 and 2–2 in equal amounts resulted in a single line in immunoelectrophoresis, not occupying the same electrophoretic position, however, as the normally occurring heterozygote *Hp* 2–1, but being more displaced towards the anode (Fig. 3). If the mixtures consist of unequal amounts of these homozygotic haptoglobin groups, varying degrees of asymmetry and displacement of the precipitates will be observed (Fig. 4).

Therefore, it seems as if the shape of the precipitates might serve as a valuable method of characterization of certain precipitating components and also that information will be obtained as to degree of 'microheterogeneity' of proteins, being at least from the serological point of view characterized as homogeneous and where also proof of heterogeneity is not obtained by conventional electrophoretic methods. Also, the electrophoretic position of precipitates might be regarded as a reflexion of the average distribution of serologically identical molecule populations in the electrophoretic field, whereas the shape of the precipitates might indicate the quantitative distribution or scatter of molecule populations in the electrophoretic field from a serological point of view.

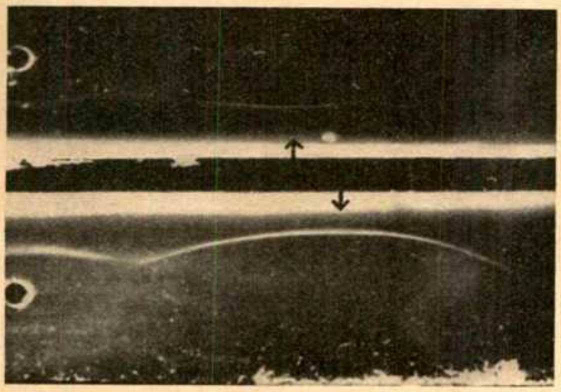

Fig. 4. Immunoelectrophoresis of mixture of *Hp* 1–1 : 0·4 ml., and *Hp* 2–2 : 0·1 ml. (upper basin) and in reversed proportions (lower basin). By means of absorption, only the haptoglobin-precipitates are visible in the α_2-globulin region

However, also other factors might affect the shape of the precipitate, for example, tailing phenomena and ratio of molecular weights between the reactants participating in the precipitate formation[11]. Finally, it must be pointed out that a serological identity-reaction might result between components which are not identical when other immune sera are used to develop the precipitates. This has been observed for some precipitating components in human sera and it seems as if different immune sera might be able to show varying degrees of reactivity with different antigenic components. In these instances an extended precipitate might be expected to show a resolution into two or more normally arc-shaped precipitates with suitable immune sera[12]. In the above-quoted examples, however, no evidence has as yet been obtained for serological differences within the haptoglobins or the other group-specific system although investigated against about 40 different immune sera prepared on rabbits and one prepared on horse.

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[1] Grabar, P., and Williams, C. A., *Biochim. Biophys. Acta*, **10**, 193 (1953).
[2] Williams, C. A., and Grabar, P., *J. Immunol.*, **74**, 158 (1955).
[3] Hirschfeld, J., *Acta Path. Microbiol. Scand.*, **46**, 229 (1959).
[4] Hirschfeld, J., *Acta Path. Microbiol. Scand.* (in the press).
[5] Grabar, P., *Thérapie*, **9**, 163 (1954).
[6] Hirschfeld, J., *Acta Path. Microbiol. Scand.*, **47**, 160 (1959).
[7] Hirschfeld, J., *Nature* (in the press).
[8] Smithies, O., *Nature*, **175**, 307 (1955).
[9] Hirschfeld, J., *Acta Path. Microbiol. Scand.*, **47**, 169 (1959).
[10] Bearn, A. G., and Franklin, E. C., *J. Exp. Med.*, **109**, 55 (1959).
[11] Korngold, L., and Leeuwen, G., *J. Immunol.*, **78**, 172 (1957).
[12] Hirschfeld, J. (to be published).

An Anthocyanin-decolorizing System in Sour Cherries

THE anthocyanins of many fruits contribute to their attractiveness and appeal. In the course of a general study of the anthocyanins we have measured the accumulation of anthocyanins together with the activity of an anthocyanin-decolorizing system in Montmorency and English Morello sour cherries[1]. Anthocyanin-decolorizing systems have previously

Table 1. ANTHOCYANIN-DECOLORIZING ABILITY AND ANTHOCYANIN ACCUMULATION IN MATURING SOUR CHERRIES

Date picked 1959	Montmorency		English Morello	
	Decolorizing ability (units/gm.)	Anthocyanin* accumulation (*O.D.* at 510 mμ)	Decolorizing ability (units/gm.)	Anthocyanin* accumulation (*O.D.* at 510 mμ)
May 29	2,740	0·515	3,200	0·875
June 9	4,950	—	5,580	—
June 19	4,680	—	6,080	—
July 2	3,790	1·80	—	—
July 9	—	—	6,950	11·0
July 15	4,075	21·9	5,560	65·4
July 24	4,220	24·2	5,310	114·0
Aug. 4	2,760	16·4	3,240	101·0
Aug. 19	3,000	14·4	2,530	113·0

* Calculated as the *O.D.* of extract from 1 gm. dry weight of cherries dissolved in 1 ml. of solution.

been reported in leaves[2-4] and these, in common with the system found in cherries, apparently involve the action of an oxidizing enzyme.

The activity of the anthocyanin-decolorizing system was assayed as described by Wagenknecht *et al.*[5] using cyanidin-3-rhamnoglucoside[6] as the substrate. The enzyme was supplied as 0·5 ml. of a homogenate containing 20 per cent wet weight of pitted cherries. The results are expressed as units per gm. dry weight of cherry tissue. Anthocyanin accumulation in the cherries was measured by heating pitted cherries in McIlvaine buffer[7], *p*H 3·4, to 71·1° C., homogenizing in a blendor, filtering and diluting the extract with buffer as needed for measurement of absorption at 510 mμ. Results were converted to optical density per ml. of extract per gm. dry weight. Cherries were harvested from late May until the middle of August.

The ability of homogenates of Montmorency and English Morello cherries to catalyse the decolorization of cyanidin-3-rhamnoglucoside increased rapidly with the age of the cherries up to a period of maximal activity and then decreased as the cherries became more mature (Table 1). The peak activity of the Montmorency sour cherries occurred about a month earlier than that found with the slower maturing English Morello variety. With both varieties the highest anthocyanin-decolorizing ability was detected when the cherries had accumulated relatively little red pigment.

The accumulation of anthocyanin (Table 1) continued throughout the experimental period in the case of the English Morello cherry, but accumulation of pigment in the Montmorency cherry reached a maximum several weeks before the experiment was ended.

In order to obtain maximum decolorization by the cherry homogenates it is necessary to add catechol to the reaction mixture[5]. In the course of the investigation reported here it was found that this was true throughout the sampling period. There was a difference between cherry varieties with regard to the amount of activity retained when catechol was not included in the reaction mixture. Upon the omission of catechol English Morello homogenates retained 33 per cent of their decolorizing activity while Montmorency homogenates retained only 20 per cent of their activity found when catechol was included. This may be due to the two varieties having different amounts of substances, such as polyphenols, which might play a part similar to that of catechol in promoting the decolorization of anthocyanin.

Relatively greater anthocyanin-decolorizing ability was found in those tissues which accumulated the greatest amount of anthocyanin. For example, the more heavily pigmented English Morello cherries were more active than the Montmorency cherries. In both varieties the activity of the skins was greater than that of the less highly pigmented flesh. It is not yet known if this relationship has any significance with respect to anthocyanin synthesis.

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[1] Journal Paper No. 1191, New York State Agricultural Experiment Station, Geneva, New York.

[2] Nagai, I., *Bot. Mag. Tokyo*, **31**, 65 (1917); *Chem. Abstr.*, **12**, 1654 (1918).

[3] Paech, K., and Eberhardt, F., *Z. Naturforsch.*, **7**,b, 664 (1952).

[4] Bayer, W., and Wegman, K., *Z. Naturforsch.*, **12**,b, 37 (1957).

[5] Wagenknecht, A. C., Scheiner, D. M., and Van Buren, J. P., *Food Tech.* (in the press).

[6] Li, K. C., and Wagenknecht, A. C., *J. Amer. Chem. Soc.*, **78**, 979 (1956).

[7] "Handbook of Chemistry and Physics", 30th edit., 1405 (Chemical Rubber Pub. Co., Cleveland, 1948).

Polysaccharide Composition of Leaves

PLUM leaves (*Prunus domestica*) contain sufficient amounts of starch, cellulose, hemicellulose, and pectic substances to recommend them for studies of the biosynthesis and possible inter-relationships of these leaf polysaccharides and their monosaccharide precursors[1]. Experiments involving photosynthesis by plum leaves in carbon dioxide labelled with carbon-14 and subsequent fractionation of the leaf polysaccharides revealed marked differences, both in the extent to which carbon-14 was incorporated into the different monosaccharides of each fraction, and also in the specific activities of each monosaccharide from different fractions[2,3]. Further investigations could be facilitated by using leaves relatively richer than plum leaves in any one polysaccharide constituent. There is little information on this point, consequently a diversity of leaves has been examined for polysaccharides (Table 1).

The leaves of wheat and gooseberry were picked in July, those of all other species were gathered during the afternoons in August and early September, and immediately immersed in ethanol. After exhaustive extraction with methanol, the leaf residues were hydrolysed (*N* sulphuric acid at 100° for 16 hr.) and, after neutralization (barium carbonate), the products examined on paper chromatograms in two solvent systems (ethyl acetate/acetic acid/water, 9 : 2 : 2; butan-1-ol/pyridine/water, 10 : 3 : 3 parts v/v)[1]. The relative amounts of the monosaccharides present in each hydrolysate were estimated visually from the size and intensity of the spots revealed on chromatograms with ammoniacal silver nitrate and *p*-anisidine hydrochloride. The presence of small amounts of acid-stable aldobiouronic acids was noted in some of the hydrolysates. Neither glucurono-1 → 4-lactone nor hexuronic acid monomethyl ether was detected, thus suggesting that galacturonic acid was the major acid component. No attempt was made to study seasonal or diurnal variations in composition, or to determine the amount of fructan, if any, or cellulose present in the leaves.

For the species so far investigated, the leaf polysaccharides were composed of a variety of monosac-

Table 1. RELATIVE AMOUNTS* OF MONOSACCHARIDES DETECTED IN LEAF POLYSACCHARIDE HYDROLYSATES

Family	Species	Galactose	Glucose	Mannose	Arabinose	Xylose	Rhamnose	Hexuronic acid
Taxaceae	*Taxus baccata* (yew)	+	+	*t*	++	*t*	—	+
Pinaceae	*Picea abies* (Norway spruce)	*t*	++	*t*	+	*t*	—	(+)
	Pinus sylvestris (Scots pine)	+(+)	++	++	++	+	*t*	(+)
	Pseudotsuga taxifolia (Douglas fir)	+	++	*t*	++	+	—	+
	Cedrus atlantica (Mt. Atlas cedar)	*t*	+++	*t*	+	*t*	—	+
Taxodiaceae	*Sequoia sempervirens* (redwood)	+	+	*t*	+	+	—	(+)
Cupressaceae	*Cupressus lawsoniana* (Lawson cypress)	+	+++	*t*	++	+	—	+
	Thuja orientalis (Chinese arbor vitae)	+	*t*	*t*	++	*t*	—	+
Fagaceae	*Fagus sylvatica* (common beech)	+	+	*t*	+	+++	—	(+)
	Quercus pedunculata (English oak)	+	+	—	++	++	—	+
Crassulaceae	*Sedum acre* (wall pepper)	+	++	—	+	*t*	—	+
Rosaceae	*Pyrus malus* (apple, var. Bramley seedling)	+	++	*t*	++	+(+)	*t*	+
	Pyrus communis (pear, var. Conference)	+	++	*t*	++	+	*t*	(+)
	Prunus domestica (plum, var. Victoria)	+	++	*t*	++	+	(+)	+
	Rubus fruticosus (cultivated bramble)	+	++	—	+	+	*t*	+
	Sorbus aria (white beam-tree)	+(+)	+	—	++	++	(+)	+
Saxifragaceae	*Ribes uva-crispa* (gooseberry)	+	++	*t*	+	(+)	(+)	++
Leguminosae	*Vicia faba* (broad bean)	+	+++	—	+	(+)	*t*	(+)
	Phaseolus vulgaris (runner bean)	+	+++	*t*	(+)	(+)	*t*	+
	Trifolium pratense (clover)	+(+)	++	—	++	+	*t*	+
Geraniaceae	*Pelargonium hortorum* (cultivated 'geranium')	+	+++	*t*	(+)	(+)	*t*	*t*
Euphorbiaceae	*Ricinus communis* (castor oil plant)	++	+++	—	+	+	*t*	+(+)
Aquifoliaceae	*Ilex aquifolium* (holly)	++	+	—	+++	+++	*t*	++
Hippo-castanaceae	*Aesculus hippocastanum* (horse chestnut)	+	+++	—	+	+	*t*	+
Araliaceae	*Hedera helix* (ivy)	+	++	*t*	+	+	*t*	+
Labiatae	*Mentha gentilis* (cultivated mint)	+	+++	*t*	(+)	(+)	*t*	+
	Salvia officinalis (cultivated sage)	+(+)	+++	*t*	++	+	*t*	+
Gramineae	*Triticum aestivum* Svenno (wheat)	*t*	+	—	+	+++	—	*t*

* In order of increasing magnitude the symbols are : — (not detected), *t* (trace), (+), +, +(+), ++, +++.

charide units, galactose, glucose, arabinose, xylose and hexuronic acid being common to them all and mannose and rhamnose frequently occurring in trace amounts. Glucose in the hydrolysates probably arose from starch which, as expected, was predominant in the leaves of many species, including cedar, bean, mint, and *Pelargonium*. Only in Scots pine needles was mannose found in more than trace amounts. Plum leaf polysaccharides were typical of the Rosaceae species examined, with roughly similar amounts of hexose and pentose components. Of particular interest for biosynthetic studies, however, were those leaves rich in pentans. For example, arabinose and xylose together formed a large part of oak and holly leaf polysaccharides, the latter apparently consisting almost entirely of xylan and the pectic substances, while xylan was a major component of wheat and beech leaves, and in yew and *Thuja orientalis* the araban component was predominant.

Wheat plants have already been used successfully to investigate xylan and cellulose biosynthesis[4], and the results presented here indicate the utility of other species for studies of polysaccharide metabolism. In this respect further studies are in progress.

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[1] Andrews, P., and Hough, L., *J. Chem. Soc.*, 4476 (1958).
[2] Hough, L., and Pridham, J. B., *Nature*, **177**, 1039 (1956).
[3] Andrews, P., and Hough, L., *Biochem. J.*, **67**, 11,*P* (1957) : *J. Chem. Soc.*, 4483 (1958).
[4] Neish, A. C., *Can. J. Biochem. Physiol.*, **36**, 187 (1958).

PHYSIOLOGY

Effect of Cytotoxic Agents on Antibody Production

IT has long been known that antibody-production may be inhibited by certain cytotoxic agents, for example, X-radiation, benzene and mustard gas[1]. More recently, the number of agents reported to have this effect has been extended to include cortisone[2], nitrogen mustards[3], colchicine[4], deoxypyridoxine[5], 8-azaguanine[6], various carcinogens[7], β-3-thienylalanine[8], 6-mercaptopurine[9] and amethopterin[10]. Many of these are potent anti-tumour compounds.

The immune response produced by the uptake and catabolism of antigen may be divided into : (1) the induction phase, usually lasting some days, during which antibody is not produced ; (2) the production phase, in which antibody protein is synthesized and released. X-rays and cortisone inhibit the immune response mainly by acting on the induction phase, although they may also affect phagocytosis. If exposure to these agents is delayed until antibody-synthesis is proceeding, their effect is diminished or absent[11]. There is evidence that mustards[12] and 6-mercaptopurine[9] behave similarly. Since these agents strongly inhibit nucleic acid synthesis, and particularly synthesis of deoxyribonucleic acid, and have much less effect on protein synthesis, these observations are relevant to the widely held belief that the induction phase is concerned with antigen-induced changes in nucleic acid metabolism[13].

With other agents inhibiting the immune response, most work has involved repeated injections of agent or antigen or both, so that the critical time of the agent's action is obscured. Such substances as β-3-thienylalanine and deoxypyridoxine, which require special dietary conditions for their action, probably inhibit the immune response largely by interfering with the production of protein, and their anti-tumour action also is probably mainly nutritional.

Table 1

Compound	Dose (mgm./kgm.)	Deaths	Titres ($\log_2$)		
			Test	Control	Difference
Group *A*					
Amethopterin	20	2/5	9	7	+ 2
8-Azaguanine	1,500	3/5	8	7	+ 1
Chlorambucil	35	0/5	8	10	− 2
Daraprim	700	3/5	4	7	− 3
6-Mercaptopurine	150	1/5	3	7	− 4
Methyl acetamide	2,750	1/5	9	8	+ 1
Methyl *bis*(β-chloroethyl)amine	2·4	2/5	8	7	+ 1
Methyl formamide	5,000	1/5	8	8	0
Ribonuclease	2,500	0/5	6	8	− 2
Stilbœstrol	1,000	3/5	7	8	− 1
Triethylene melamine	2	2/5	5	8	− 3
Urethane	1,250	1/5	9	8	+ 1
Group *B*					
8-Azaxanthine	3,000	3/5	7	8	− 1
Benzimidazole	600	3/5	9	8	+ 1
Boric acid	2,500	3/5	9	8	+ 1
Caffeine	200	1/5	8	8	0
Deoxypyridoxine	275	1/5	8	8	0
D-Glucosamine	8,000	3/5	7	8	− 1
Methyl carbamate	1,000	1/5	6	7	− 1
Ovalbumin (hen)	2,500	0/5	7	7	0
Potassium cyanide	7·5	1/5	8	8	0
Serum albumin (bovine)	2,500	0/5	8	7	+ 1
Streptomycin sulphate	750	2/5	8	7	− 1
β-2-Thienylalanine	5,000	1/5	9	8	+ 1

It was thought that investigations along these lines might throw further light both on the initiation of the immune response and on the mode of action of anti-tumour compounds. A wide range of substances was therefore examined, by administering simultaneously single doses of substance and antigen, and titrating antibody 10 days after the injection. By this timing it was hoped to identify substances that interfere with the induction phase of the immune response and do not act simply by impairing protein synthesis.

Male *A2G* mice, weighing 17–25 gm., were given single subcutaneous injections of the compounds studied and, immediately afterwards, 0·2 ml. T.A.B. vaccine intraperitoneally. The compounds were dissolved or suspended in 0·85 per cent saline or, when appropriate, in arachis oil. Controls received 1 ml. saline or arachis oil. Ten days later serum from groups of 5 mice was pooled and titrated for *Salmonella typhi H*-agglutinins. Titres are expressed in Table 1 as logarithms to the base 2, and a reduction of 2 in titre was regarded as significant evidence of inhibition.

Each compound was tested at several dose-levels and, except for the proteins, which present difficulties in this respect, the range included doses that killed a proportion of the animals. Table 1 gives only the results obtained at the higher end of this range. Published toxicity-levels were only a rough guide to dosage, owing partly to concurrent administration of vaccine. Colchicine, for example, could not be tested satisfactorily; its *LD*50, normally 1·75 mgm./kgm. in these mice, was 0·35 mgm./kgm. when the vaccine was given simultaneously.

In Table 1 group *A* consists of compounds that are used clinically against human tumours, leukæmia or polycythæmia vera or that are undoubtedly effective against transplanted animal tumours without requiring special dietary conditions. Group *B* are not effective antitumour agents as judged by these criteria.

Of the 24 compounds, five significantly reduced antibody-production. All were in group *A*. Four of these are clinically useful in human malignant disease. The fifth, ribonuclease, is said to be active against various mouse tumours[14], but there has been no published report of its effect on human neoplasms. No compound ineffective against tumours ever caused significant impairment of antibody-production under the conditions of this experiment, although most of them were tested almost to the limit of tolerance.

Some of the group *A* compounds inhibited antibody-production in non-lethal doses, whereas group *B* included compounds that were ineffective in doses that killed most of the animals. It is therefore unlikely that the inhibition of the immune response was due to general toxicity. In fact, the doses were such that the same proportion of deaths occurred in both groups.

Ribonuclease appears of special interest, but, since it is itself an antigen, the possibility that it acts here by 'antigenic competition' cannot yet be excluded, although the other proteins tested failed to inhibit antibody production. Deoxyribonuclease, 1,000 mgm./kgm., had no effect on the antibody titre.

These preliminary results suggest that this experimental system may provide a useful adjunct to other methods for screening anti-tumour compounds.

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[1] Hektoen, L., *J. Infect. Dis.*, **17**, 415 (1915); **19**, 69 (1916). Hektoen, L., and Corper, H. J., *ibid.*, **28**, 279 (1921).

[2] Kass, E. H., and Finland, M., "Ann. Rev. Microbiol.", **7**, 361 (1953).

[3] Spurr, C. L., *Proc. Soc. Exp. Biol.*, **64**, 259 (1947). Phillips, F. S., Hopkins, F. H., and Freeman, M. L. H., *J. Immunol.*, **55**, 289 (1947).

[4] Forman, C., Seifter, J., and Ehrich, W. E., *J. Allergy*, **20**, 273 (1949). Fagraeus, A., and Gormsen, H., *Acta Path. Micro. Scand.*, **33**, 421 (1953).

[5] Stoerck, H. C., *Ann. N.Y. Acad. Sci.*, **52**, 1302 (1950).

[6] Malmgren, R. A., Bennison, B. E., and McKinley, T. W., *J. Nat. Cancer Inst.*, **12**, 807 (1952).

[7] Malmgren, R. A., Bennison, B. E., and McKinley, T. W., *Proc. Soc. Exp. Biol. Med.*, **79**, 484 (1952).

[8] Wissler, R. W., Frazier, L. F., Soules, K. H., Barker, P., and Bristow, E. C., *Arch. Path.*, **62**, 62 (1956).

[9] Schwartz, R., Stack, J., and Damashek, W., *Proc. Soc. Exp. Biol. Med.*, **99**, 164 (1958).

[10] Uphoff, D., *Proc. Soc. Exp. Biol. Med.*, **99**, 651 (1958).

[11] Fagraeus, A., *Acta Path. Micro. Scand.*, Supp. 93, **20** (1952). Berglund, K., *ibid.*, **38**, 311 (1956). Taliaferro, W. M., *Ann. N.Y. Acad. Sci.*, **69**, 745 (1957).

[12] Shamaeva, E. M., and Pankova, S. S., *J. Microbiol. Epidemiol. Immunobiol.*, **28**, 489 (1957). Green, D. M., *Brit. J. Exp. Path.*, **39**, 192 (1958)

[13] Burnet, F. M., "Enzyme, Antigen and Virus" (Cambridge, 1956). Schweet, R. S., and Owen, R. D., *J. Cell Comp. Physiol.*, **50**, Supp. 1, 199 (1957). Schultz, J., *Science*, **129**, 937 (1959).

[14] Ledoux, L., and Baltus, E., *Exper.*, **10**, 500 (1954). Ledoux, L., *Nature*, **176**, 36 (1955).

Influence of Insulin on the Incorporation of 2-^{14}C-Sodium Pyruvate into Glyceride Glycerol in Diabetic and Normal Baboons

DEPANCREATIZED baboons, deprived of insulin for 4 days, develop a lipæmia and a fatty liver, which may contain 34 per cent of fat when estimated on a wet-weight basis[1,2]. The lipæmia in diabetic rats has generally been attributed to mobilization of fat from the depots. In the baboon, the severity of the lipæmia and the degree of fat accumulation in the liver, particularly in young animals with poorly developed fat depots, suggested that the fat was not mobilized only from the depots, and this warranted a re-investigation of the origin of the fat in the liver and in the plasma. In this communication it is proposed to report on the incorporation of 2-^{14}C sodium pyruvate into the glyceride glycerol in a normal baboon and in depancreatized baboons either receiving insulin or deprived of insulin.

Five animals were used in this study. General data on the animals are given in Table 1.

Table 1. DATA ON EXPERIMENTAL BABOONS

Baboon	Age (yr.)	Weight (kgm.)	Sex	Treatment	Duration of pancreatectomy
440	± 6–7	14	F	Normal	—
422	± 6	12·7	F	Depancreatized on insulin	7 months
355	6	20	M	Depancreatized without insulin	2½ years
448	± 3	11	M	Depancreatized without insulin	2½ months
403	3½	8·1	M	Depancreatized without insulin	9 months

During the course of the experiment, baboons 355, 448 and 403 received no insulin for 96 hr. Each animal received a total of 500 μc. of 2-^{14}C sodium pyruvate (supplied by the Radiochemical Centre, Amersham) in three divided doses injected intravenously as a saline solution over a period of three days. 6 hr. after the last pyruvate injection blood was drawn for the isolation of serum lipids ; at the same time, samples of liver and depot fat were removed and extracted for fats. The triglyceride fractions of the fats were separated by chromatography on silicic acid. The triglycerides were hydrolysed by boiling with alcoholic potassium hydroxide. After extraction of the fatty acids with ether, the glycerol was isolated from the aqueous fraction as glycerol tribenzoate. The substance was recrystallized from methanol and the purity of the material was checked by melting point determination. A further check on the purity of the material was obtained from the wet combustion to carbon dioxide according to the method of van Slyke, Plazin and Weisiger[3]. All samples were counted as barium carbonate with a proportional gas counter. Details of the method used for plating and counting will be published shortly.

The results of the incorporation of carbon-14 into the glyceride glycerol of serum, liver and depot fat in the five baboons are shown in Table 2. We have demonstrated that it is possible to calculate, with a reasonable degree of accuracy, the specific activity of the glycerol from the observed total activity of the unhydrolysed triglyceride fraction after deduction of the activity due to the fatty acids. As limited amounts of triglycerides were obtained from the serum of baboons 422 and 440, we have no information on the radioactivity incorporated into the glycerol in these cases. However, the evidence is very suggestive that the levels in the serum reflect those in the liver and we suspect that the radioactivity in the glyceride glycerol of the serum in these two baboons was low.

Table 2. SPECIFIC ACTIVITIES OF GLYCERIDE GLYCEROL OF SERUM, LIVER AND DEPOT FAT (COUNTS/MIN./M*M*) OF DEPANCREATIZED BABOONS AFTER ADMINISTRATION OF 2-^{14}C SODIUM PYRUVATE

Baboon No.	Treatment	Serum	Liver	Depot fat
355	Depancreatized without insulin	62,916*	73,268* 68,632	3,956* 2,484
448	Depancreatized without insulin	—	99,544	13,064
403	Depancreatized without insulin	120,888* 113,449	111,504*	6,532
422	Depancreatized on insulin	—	0	0
440	Normal	—	0	0

* Calculated.

Attention is directed to the following experimental findings : (1) Carbon-14 is markedly incorporated into the glyceride glycerol of the liver and plasma and to a much lesser extent into the glycerol of the fat depots (Nos. 355, 448 and 403, Table 2). (2) The administration of insulin to the depancreatized baboon completely blocks the incorporation of carbon-14 into the glycerol of liver and depot fat (No. 422, Table 2). (3) Incorporation does not occur in the normal baboon (No. 440, Table 2). (4) The extent of the incorporation is the same in the glyceride glycerol in the serum and in the liver of depancreatized insulin-deprived baboons, as determined by experiment or by calculation (Nos. 355 and 403). (5) The extent of incorporation is from 30 (No. 355) to 200 times greater (No. 403) in the liver glycerol than in the fat depots.

These observations, taken in conjunction with others relating to the fatty acids (unpublished results), would seem to indicate that if fat is mobilized from the depots it is not mobilized as triglycerides of the depots but is hydrolysed before entering the circulation. Furthermore, the close correspondence between the glycerol in the liver and serum would suggest that the triglycerides of the serum are derived from the liver and not primarily from the depots. The blocking effect of insulin on the synthesis of glycerol from pyruvate or from metabolites derived from pyruvate is a noteworthy phenomenon.

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[1] Gillman, J., and Gilbert, C., *Brit. Med. J.*, i, 57 (1958). Gillman, J., Gilbert C., Epstein, E., and Allan, J. C., *Brit. Med. J.*, ii, 1260 (1958).

[2] Savage, N., Gilbert, C., and Gillman, J. (in the press).

[3] van Slyke, D. D., Plazin, J., and Weisiger, J. R., *J. Biol. Chem.*, 191, 299 (1951).

Action of Thyroxine Analogues on Frog Melanocytes

THE melanocyte-stimulating hormones and caffeine darken frog melanocytes by causing dispersion of melanin granules throughout the cell. While the end effect of these two substances on melanocyte opacity is the same, there are several differences in the way melanocyte-stimulating hormone and caffeine bring about dispersion. Lightening of melanocytes is produced by aggregation of melanin granules towards the centre of the cell. We found that triiodothyronine was an effective lightening agent of frog skin previously darkened by melanocyte-stimulating hormone but had no action on caffeine-treated skin. Because of this result we decided to test thyroxine and triiodothyroacetic acid for lightening properties.

Skin from the legs and thighs of eight frogs (*Rana pipiens*) was mounted on rings[1] and arranged in four comparable groups as follows:

Group *A*	LLg1	LTh2	RTh3	RLg4	LLg5	LTh6	RTh7	RLg8
Group *B*	RLg1	LLg2	LTh3	RTh4	RLg5	LLg6	LTh7	RTh8
Group *C*	RTh1	RLg2	LLg3	LTh4	RTh5	RLg6	LLg7	LTh8
Group *D*	LTh1	RTh2	RLg3	LLg4	LTh5	RTh6	RLg7	LLg8

L, left; R, right; Lg, leg; Th, thigh.

Before any observations were made, all skin specimens were soaked in four changes of frog Ringer's solution for 2 hr. Each specimen was maintained in 20 ml. of Ringer's solution throughout the experiment. Changes in the skin were measured with a photoelectric reflexion meter[1]. The initial reading of each sample was taken as its individual base-line, and increase or decrease in reflectance was measured from this.

Two groups were treated with 7·5 units of melanocyte-stimulating hormone per 20 ml. Ringer's solution, and two groups were treated with 20 mgm. of caffeine per 20 ml. Ringer's solution. These quantities of melanocyte-stimulating hormone and caffeine produced approximately the same degree of darkening. The melanocyte-stimulating hormone had been made from bovine posterior pituitary powder[1].

Pronounced darkening occurred in all groups and was followed as a decrease in reflectance for 60 min. At this time sodium triiodothyronine was added to one group treated with melanocyte-stimulating hormone and to one group treated with caffeine. The final concentration of the hormone was 5 μgm. per ml. Changes in reflectance were followed for 50 min. more. As shown in Fig. 1, marked lightening occurred in the group treated with melanocyte-stimulating hormone following the addition of triiodothyronine. There was no change in the group treated with caffeine.

The experiment was repeated using thyroxine as the sodium pentahydrate salt and triiodothyroacetic acid as the diethanolamine salt. Again marked lightening occurred in the groups previously treated with melanocyte-stimulating hormone but not with those treated with caffeine.

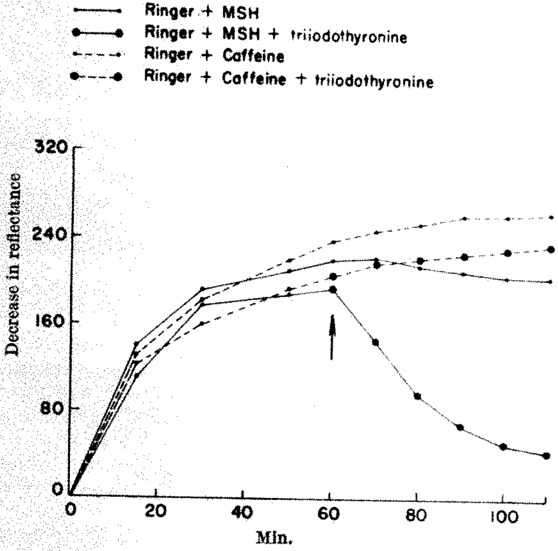

Fig. 1. The effect of triiodothyronine on melanocyte-stimulating hormone- and caffeine-darkened frog skin. Four comparable groups of skins were darkened, two with melanocyte-stimulating hormone and two with caffeine. Decrease in reflectance was followed. After 60 min. triiodothyronine (5 μgm./ml.) was added to one melanocyte-stimulating hormone- and to one caffeine-treated group. Triiodothyronine reversed the darkening action of melanocyte-stimulating hormone but not that of caffeine

The minimum amount required to initiate lightening was 0·5 μgm. per ml. of sodium triiodothyronine, 1·5 μgm. per ml. of thyroxine sodium pentahydrate and 25 μgm. per ml. of the diethanolamine salt of triiodothyroacetic acid. These potency ratios are in approximate agreement with those representing the thyroid hormone actions of these compounds as measured by inhibition of iodine uptake[2].

Melatonin, noradrenaline, serotonin and hydrocortisone can reverse the darkening actions of both melanocyte-stimulating hormone and caffeine. The only compounds known to reverse the melanocyte-stimulating hormone effect but not that of caffeine are acetylcholine and thyroxine and its two analogues[3].

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[1] Shizume, K., Lerner, A. B., and Fitzpatrick, T. B., *Endocrinol.*, **54**, 553 (1954).

[2] Money, W. L., Meltzer, R. I., Feldman, D., and Rawson, R. W., *Endocrinol.*, **64**, 123 (1959).

[3] Wright, M. R., and Lerner, A. B., *Endocrinology* (in the press).

Immunochemical Observations on Antisera to Purified Human Growth Hormone

RECENTLY, Hayashida and Li[1] and Read[2] have reported that purified preparations of human growth hormone were antigenic when injected with adjuvants into rabbits. Serum antibodies were demonstrated by the precipitin ring test[1] and by a modification[2] of the tanned cell hæmagglutination method of Boyden[3]. It was also shown that the rabbit antiserum inhibited the biological activity of human growth hormone[1]. Read[2] has employed rabbit antiserum to measure the concentration of growth hormone in human serum by the hæmagglutination–inhibition reaction[3,4]. It seemed essential in assessing the specificity of this method to investigate the purity of the antigen and the specificity of the antisera. In this report immunochemical evidence is presented which confirms that human growth hormone is a potent antigen. In addition, results were obtained which indicate that the Raben preparation of human growth hormone[5] is a highly purified antigen.

A relatively potent antiserum was prepared by the injection of rabbits with human growth hormone prepared by Raben (*Homo* No. 6) according to the following schedule[1]: 2 mgm. of human growth hormone in complete Freund's adjuvant (Difco) was injected both subcutaneously and intraperitoneally on the first and third weeks; on the fifth week human growth hormone was suspended in 1 per cent potassium alum[6] and 1 mgm. was injected by both the subcutaneous and intraperitoneal routes. The total amount of antigen was 10 mgm. per rabbit. The individual rabbit antisera were preserved by the addition of 1 per cent merthiolate to a final concentration of 1 : 20,000 after passage through a 'Millipore' filter.

The immunological specificity of the reactants was studied (1) by passive hæmagglutination and the hæmagglutination–inhibition reaction using tanned sheep erythrocytes conjugated with human growth hormone[2]; (2) by the double diffusion gel plate technique of Ouchterlony as modified by Halbert *et al.*[7]; (3) by the Preer micromethod[8] for double diffusion in agar; (4) by Scheidegger's micromodification[9] of the immuno-electrophoresis procedure of Grabar and Williams; and (5) by the quantitative precipitin method, the nitrogen content of the precipitates being determined by the Markham micro-Kjeldahl method[6]. Human growth hormone from the same lot (*Homo* No. 6) was employed in experiments in Read's and in our laboratories.

The passive hæmagglutination titres of individual rabbit antisera ranged from 1/6,400 to 1/51,200. As little as 1·35 mμgm. nitrogen of antigen was detectable by Read's modification of the hæmagglutination-inhibition reaction. A single band of precipitation was obtained in Ouchterlony plates when undiluted antiserum was tested against dilutions of human growth hormone of 1 mgm./ml. to 10^{-2} mgm./ml. The intense band was completely removed following absorption of antiserum with human growth hormone. Complete fusion of single bands was demonstrated when antiserum was set up against 500 μgm./ml. dilutions of Raben's human growth hormone and Li's human growth hormone[10], a crude preparation of human pituitary, and simian growth hormone. No spur formation to indicate partial identity was found. A band of precipitation was not detected in the agar plates against 2 mgm./ml. of human adrenocorticotrophic hormone (Organon), bovine growth hormone, human chorionic and menopausal gonadotropins, or samples of unabsorbed human serum. Similarly, in studies of the antisera and of purified human growth hormone and crude pituitary extract, and of simian growth hormone by the Preer method, a single disk-like band of precipitation was obtained; the equivalence point was noted at a half dilution of antiserum against 62·5 μgm./ml. of human growth hormone (Fig. 1).

Investigation of the reactants by the immuno-electrophoresis procedure showed, in experiments in which the anti-human growth hormone serum was added to a longitudinal central trough and 500 μgm./ml. of human growth hormone to one lateral well and 1 mgm./ml. of a crude pituitary extract to the other lateral well, a single arc of precipitation against each preparation (Fig. 2, bottom). In this system human growth hormone exhibited the relative mobility of a slow β-globulin. The antigen did not react with horse hyperimmune anti-human serum (Fig. 2, top) or rabbit anti-human globulin. These studies indicate that contamination of purified human growth hormone with serum proteins is negligible. Since antibodies to human growth hormone were not detected in the Paris anti-human serum, it is quite reasonable to assume that the amount of human growth hormone contained in normal human serum is insufficient to elicit antibodies in the horse. The lack of cross-reaction between human growth hormone antiserum and human adrenocorticotrophic hormone (2 mgm./ml.), bovine growth hormone (1 mgm./ml.), and several human sera was confirmed by immuno-electrophoresis. Interestingly, the single, smooth, narrow arc of precipitation found with simian growth hormone revealed a slightly different electrophoretic mobility than did that obtained with human growth hormone, being situated slightly closer to the cathode.

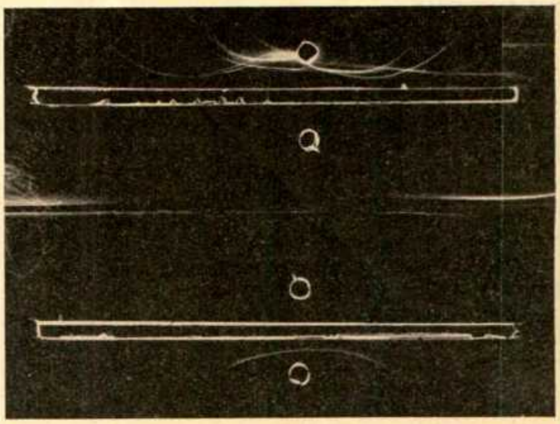

Fig. 2. Top, immuno-electrophoretic precipitin pattern of human sera (upper well) with horse hyperimmune anti-human serum (Paris serum No. 491). No reaction noted between human growth hormone (lower well) and 'Paris serum' (longitudinal trough). Bottom, immuno-electrophoretic precipitin pattern of human growth hormone (500 μgm./ml.) (lower well) with rabbit antiserum (longitudinal trough). No reaction of antiserum with human adrenocorticotrophic hormone (2 mgm./ml.) (upper well)

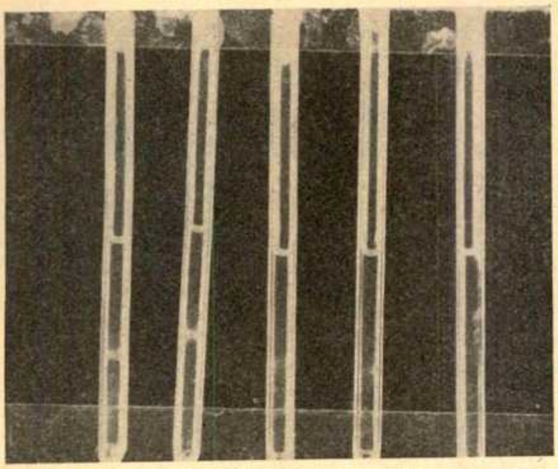

Fig. 1. Preer tubes with rabbit anti-human growth hormone serum in dilutions (from left to right) of undiluted, $\frac{1}{2}$, $\frac{1}{4}$, $\frac{1}{8}$ and $\frac{1}{16}$ in which the concentration of growth hormone added to the tubes was kept constant at 62·5 μgm./ml. Equivalence point at $\frac{1}{2}$ dilution of antiserum with single band of precipitation

The quantitative precipitin method was used to study the total amount of antigen–antibody precipitated by the antiserum after incubation with varying concentrations of antigen. Preliminary results indicate a precipitin curve consistent with a single antigen–antibody system.

It was also shown that the injection of the same antisera used in the studies described above and human growth hormone by separate routes into hypophysectomized rats completely inhibited the biological activity of the hormone, as assayed by weight gain and by the tibia test.

The results of these experiments indicate that the Raben preparation of human growth hormone is a potent antigen with a high degree of immunological homogeneity. Antisera prepared by injection of human growth hormone into rabbits could not be shown to contain precipitins or other antibodies specifically related to human antigens other than human growth hormone. Evidence was obtained of only a single antigen–antibody system. It seems almost certain from the results that the single system observed actually represents the human growth hormone–anti-human growth hormone reaction. However, the limitations of the immunochemical methods do not exclude the possibility that this preparation of human growth hormone (*Homo* No. 6) is contaminated with trace amounts of other antigens or with weak antigens or, of course, with non-antigenic material.

The physicochemical and structural characteristics of simian and human growth hormone show important similarities[11] and both hormones exhibit biological activity in man. The different electrophoretic mobility of these two antigens was clearly shown by immuno-electrophoresis.

The apparent immunological specificity of purified human growth hormone and its antibodies strongly supports the usefulness of the hæmagglutination-inhibition reaction as a specific method for the quantitative estimation of growth hormone. Evaluation of the Read method as applied to human sera is in progress in this laboratory.

Note added in proof. In additional experiments in which human adrenocorticotrophin prepared by M. S. Raben was used, this material was found to be contaminated with small amounts of human growth hormone, in contrast to the highly purified Organon preparation employed in previous studies.

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[1] Hayashida, T., and Li, C. H., *Science*, **128**, 1276 (1958).

[2] Read, C. H., and Stone, D. B., *A. Med. Assoc. J. Dis. Child.*, **96**, 538 (1958). Read, C. H., in "Progress in Clinical Endocrinology", Astwood, E. B. (ed.), **2** (Grune and Stratton, Inc., New York; in the press).

[3] Boyden, S. V., *J. Exp. Med.*, **93**, 107 (1951).

[4] Stavitsky, A. B., *J. Immunol.*, **72**, 360 (1954).

[5] Raben, M. S., *Science*, **125**, 883 (1957).

[6] Kabat, E., and Mayer, M., "Experimental Immunochemistry" (Charles C. Thomas, Springfield, Ill., 1948).

[7] Halbert, S., Swick, L., and Sonn, C., *J. Exp. Med.*, **101**, 557 (1958).

[8] Preer, jun., J., *J. Immunol.*, **77**, 52 (1956).

[9] Scheidegger, J. J., *Intern. Arch. Allergy*, **7**, 103 (1955).

[10] Li, C. H, and Papkoff, H, *Science*, **124**, 1293 (1956).

[11] Léonis, J., and Li, C. H., *Biochim. Biophys. Acta*, **28**, 520 (1958).

Effect of Heavy Water on Different Types of Muscle and on Glycerol-extracted Psoas Fibres

On considering the suggestion that more attention be directed to the role of water in biochemical reactions[1], the possibility was entertained that by using heavy water further information might be obtained about physical-chemical processes in muscle. An investigation was therefore undertaken to observe the effects of heavy water on heart, intestinal, uterine and voluntary muscle.

Concentrations of heavy water between 99·8 and 25 per cent decreased the force of contraction and the rate of the spontaneously beating, isolated atria of the frog (*Xenopus laevis*). Similar effects have been observed previously on the frog heart[2]. The onset of the effects on the atria of *Xenopus* was noted 50 sec. after immersion in heavy water-Ringer solution. With 99·8 per cent heavy water, the force decreased by 90 per cent of its basal value after 2½ min. In most instances the atria either stopped beating or slowed down to one impulse every 5 or 7 min. after being immersed for about 20 min. in heavy water. The velocity of contraction was reduced[3], as was also the velocity of relaxation (Fig. 1).

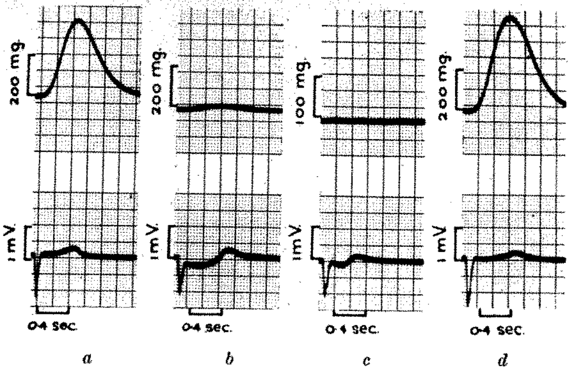

Fig. 1. Simultaneous recordings of tension (upper tracing) obtained with an RCA transducer and unipolar surface electroatriogram (lower tracing). *a*, Basal recording in normal water–Ringer; *b*, after 5 min. in 99·8 per cent heavy water–Ringer; *c*, after 17 min. in 99·8 per cent heavy water–Ringer (note no recordable contraction with increase in gain of amplifier); *d*, recovery after 15 min. in normal water–Ringer (note that the force of contraction is greater than in the basal recording)

Simultaneous recordings of the surface electroatriogram (Fig. 1) revealed that 99·8 per cent heavy water reduced the amplitude of the depolarization wave by about 13 per cent on the average. On the other hand, the amplitude of the repolarization wave was increased after 5 min. and then decreased towards the basal value; changes in the configuration of the repolarization wave were obvious. The duration of depolarization and repolarization increased by approximately 40 and 60 per cent, respectively. The interval between the onset of the electrical impulse and the contraction increased by about 90 per cent of the basal value. These manifestations in the mechanical and electrical parameters were reversed after transferring the atria to normal water-Ringer solution.

A particularly interesting feature was the dissociation in response of the electrical and contractile events. In 4 out of 10 experiments the contraction was not detectable while the impulse continued to be propagated (Fig. 1). Hodgkin and Horowicz produced similar dissociation of the twitch and action potential of single muscle fibres with high concentrations of sodium chloride[4].

After these preliminary experiments, attempts were made to elucidate the sites where heavy water affected the contraction of muscle. First, electrical stimulation of the heavy water-inhibited atria resulted in an action potential. The contractile response was, however, small and never greater in force than that seen prior to the arrest of the heart. It appears, therefore, that the membrane of the muscle fibres of the atria remained excitable while the spontaneous impulse was suppressed by inhibition of the pacemaker by heavy water.

Secondly, chemical stimulants such as adrenalin, noradrenalin and veratrine increased the force of contraction of atria treated with 75 per cent heavy water. In passing, it may be mentioned that heavy water also diminished the force of contraction of isolated rabbit duodenum. However, 75 per cent heavy water did not modify the effects of acetylcholine on rabbit duodenum and of oxytocin on the guinea pig's uterus. A high concentration of calcium chloride (4·5 m*M*), sufficient approximately to double the force of contraction in untreated atria, had no inotropic effect on atria treated with heavy water. Whereas in untreated atria both adrenalin and noradrenalin increased the rate of rhythmic contraction, this chronotropic effect did not occur in the presence of heavy water.

After establishing that heavy water decreased markedly the force of contraction of voluntary muscle (rectus abdominis of frog and rabbit psoas), a third series of investigations was carried out with heavy water on psoas muscle fibres extracted with glycerol[5]. In the case of these glycerol-extracted fibres, however, heavy water did not retard the degree of shortening induced by adenosine triphosphate. To increase the probability of deuterium exchange, living fibres were activated by electrical stimulation while submerged in heavy water and then extracted in 99·8 per cent heavy water at 4° C. instead of in 50 per cent glycerol. The response of the fibres treated in this way was again not affected by adenosine triphosphate dissolved in heavy water–salt solution. Further experiments on the glycerol-treated fibres with other nucleotides revealed that heavy water reduced the amount of shortening produced by inosine triphosphate and uridine triphosphate but not the amount produced by adenosine triphosphate and cytodine triphosphate (Fig. 2). In so far that the difference in molecular structure between inosine triphosphate and adenosine triphosphate on one hand and between uridine triphosphate and cytodine triphosphate on the other is in the 6th position of the purine and the pyrimidine rings respectively, it is suggested that reactions involving the 6th position of either inosine triphosphate or uridine triphosphate were retarded by deuterium.

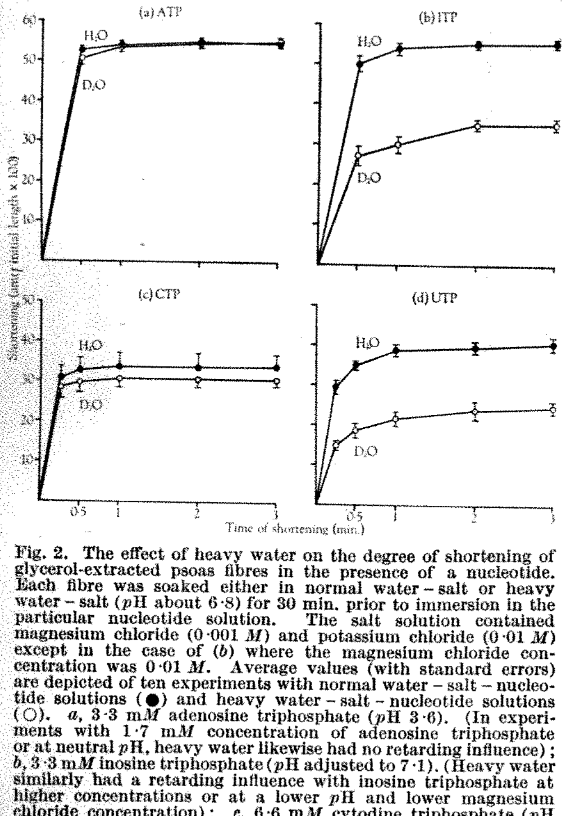

Fig. 2. The effect of heavy water on the degree of shortening of glycerol-extracted psoas fibres in the presence of a nucleotide. Each fibre was soaked either in normal water – salt or heavy water – salt (*p*H about 6·8) for 30 min. prior to immersion in the particular nucleotide solution. The salt solution contained magnesium chloride (0·001 *M*) and potassium chloride (0·01 *M*) except in the case of (*b*) where the magnesium chloride concentration was 0·01 *M*. Average values (with standard errors) are depicted of ten experiments with normal water – salt – nucleotide solutions (●) and heavy water – salt – nucleotide solutions (○). *a*, 3·3 m*M* adenosine triphosphate (*p*H 3·6). (In experiments with 1·7 m*M* concentration of adenosine triphosphate or at neutral *p*H, heavy water likewise had no retarding influence); *b*, 3·3 m*M* inosine triphosphate (*p*H adjusted to 7·1). (Heavy water similarly had a retarding influence with inosine triphosphate at higher concentrations or at a lower *p*H and lower magnesium chloride concentration); *c*, 6·6 m*M* cytodine triphosphate (*p*H 4·35); *d*, 6·6 m*M* uridine triphosphate (*p*H 3·56). (Insufficient material precluded studies on uridine triphosphate and cytodine triphosphate at varying concentrations and *p*H's)

As the degree of shortening of the contractile proteins was not necessarily reduced by heavy water, the possibility exists that either the substitution of deuterium for hydrogen in the contractile proteins did not in itself retard the response or that little exchange took place. Taken in conjunction with the observed dissociation of the electrical and contractile events in the living muscle, it seems likely that the energy transfer mechanism between the membrane and the contractile protein is retarded or blocked by the isotopic effect of deuterium. The response produced by the drugs, for example adrenalin, could be due to the activation of the energy transfer process. Calcium ions, on the other hand, could not counteract the effects of heavy water. The manner whereby heavy water inhibited the pacemaker remains to be elucidated.

It thus appears that heavy water can be useful as a tool in the dissection of the physical-chemical processes of muscle contraction. Full details of the experiments will be published elsewhere.

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[1] Szent-Györgyi, A., "Chemical Physiology of Contraction in Body and Heart Muscle" (Academic Press, New York, 1953).

[2] Barnes, T. C., and Warren, J., *Science*, **81**, 346 (1935). Brandt, W., *Klin. Wschr.*, **14**, 1597 (1935). Verzar, F., and Haffter, C., *Pflüg. Arch. ges. Physiol.*, **236**, 714 (1935).

[3] Goodall, M. C., *Nature*, **182**, 677 (1958).

[4] Hodgkin, A. L., and Horowicz, P., *J. Physiol.*, **136**, 17p (1957).

[5] Szent-Györgyi, A., *Biol. Bull.*, **96**, 140 (1949).

Effect of Parathyroid Extract on Citric Acid Concentration in the Rat Kidney

THE influence of parathormone on the level of citric acid in plasma was first mentioned by Sjöström[1], who found an elevated level of citric acid in patients with hyperparathyroidism; this finding was afterwards confirmed by other authors. The effect of parathormone on the citric acid level was verified by Neuman *et al.*[2], who measured the amount of the acid in the venous blood of bones and found an elevated level after the application of parathormone. The purpose of the work reported in this communication was to prove this effect of parathormone on the metabolism of citric acid also in other tissues and to try to explain the mode of action.

The concentration of citric acid in the skeleton and kidneys of female rats, 150–160 gm., was observed. The rats received 10 U. of parathyroid extract ('Parathyroid', Paines and Byrne) intramuscularly, and were killed at intervals of 1–24 hr. after the injection. The kidneys were immediately homogenized in liquid air; the dissected left femur was extracted for 7 days in a solution of 10 per cent trichloracetic acid. Citric acid was determined by Gey's method[3]. In order to explain the mode of action of parathormone, the parathyroid extract was given

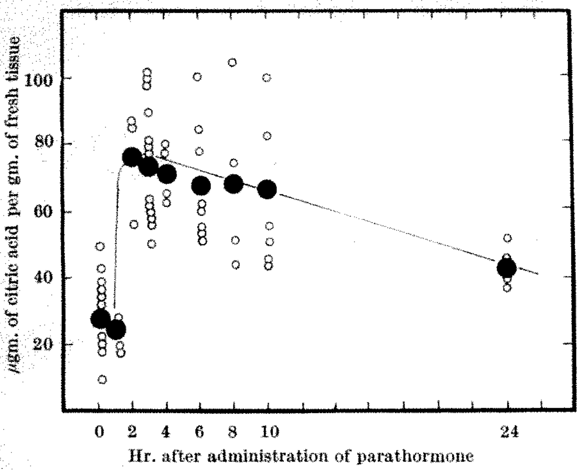

Fig. 1

3 hr., and a freshly prepared sodium fluoracetate solution (5 mgm./kgm. body-weight) was given 1 hr., before the animal was killed, to obtain the maximal effect of both compounds.

The results are presented in Table 1 and Fig. 1. The extract of parathyroid glands raises sharply the concentration of citric acid in the kidneys after latent periods of 60 min. The elevation of citric acid is statistically significant ($P \leqslant 0{\cdot}01$) in comparison with the control group. It reaches its maximum in 2–3 hr. after the injection. The concentration of citric acid remains at approximately the same level up to 10 hr. after the injection and returns to about the starting concentration after 24 hr.

Table 1. CONCENTRATIONS OF CITRIC ACID MGM./GM. OF FRESH TISSUE IN DIFFERENT TIME INTERVALS AFTER THE APPLICATION OF PARATHYROID EXTRACT

Control	1 hr.	2 hr.	3 hr.	4 hr.	6 hr.	8 hr.	10 hr.	24 hr.
0·019	0·030	0·076	0·10	0·065	0·060	0·044	0·096	0·044
0·023	0·028	0·085	0·10	0·080	0·054	0·074	0·082	0·050
0·039	0·018	0·085	0·080	0·062	0·051	0·104	0·099	0·038
0·028	0·018	0·056	0·10	0·078	0·055	0·051	0·068	0·038
0·050			0·060		0·083		0·043	
0·043			0·080		0·10		0·056	
0·030			0·080		0·078		0·045	
0·010			0·050		0·061		0·050	
0·020			0·060					
0·033			0·090					
0·038			0·078					
0·021			0·056					
0·037			0·061					
0·035			0·063					
			0·059					
Mean values 0·030	0·023	0·075	0·074	0·071	0·068	0·068	0·067	0·042

After the application of sodium fluoracetate, which inhibits the further degradation of citric acid in the Krebs cycle, the concentration of citric acid in the kidneys rises fifty times[4] within 1 hr. When sodium fluoracetate and parathormone were given so as to achieve the maximum effect of both compounds simultaneously, there was statistically no significant difference in the content of citric acid in the kidneys compared with the control group ($P = 0{\cdot}1$).

The content of citric acid in bones remained the same after administration of parathormone alone or in combination with fluoracetate; there was no significant difference between these groups.

Our findings point to the fact that parathormone influences the metabolism of citric acid not only in the bones but also in other tissues—in our case in the kidneys of rats. We assume that the parathormone may interfere directly with the Krebs cycle and that it may act either by raising the synthesis of citric acid or by inhibiting its degradation. The blocking of citric acid metabolism in the Krebs cycle by fluoracetate did not enhance the effect of the parathormone on the concentration of citric acid. It appears, therefore, that the interference of parathormone with the metabolism of citric acid is caused rather by inhibition of its further degradation than by augmentation of its synthesis. Similar effect has been described in the mode of action of vitamin D[5]. As, however, the quantity of citric acid is much higher after administration of fluoracetate than after parathormone, it should be taken into account that a raised synthesis of citric acid, owing to the effect of parathormone, can be hidden in the great excess of citric acid and that the relatively small changes induced by the parathormone are scarcely detectable by the method used. The same explanation can be applied to the negative response of bones to the administration of parathormone and fluoracetate because of high levels of citric acid in bones under normal conditions.

The question of the mode of interference of parathormone with the Krebs cycle is not fully explained by our findings and requires further investigation.

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[1] Sjöström, P., *Acta chir. Scand.*, Supp. 79 (1937).
[2] Neuman, W. F., Firschein, H., Chen, P. S., Mulryan, B. J., and Di Stefano, V. J., *J. Amer. Chem. Soc.*, **78**, 3836 (1956).
[3] Gey, K. F., *Intern. Z. Vitaminforsch.*, **25**, 21 (1953).
[4] Lindenbaum, A., White, M. R., and Schubert, J., *J. Biol. Chem.*, **190**, 585 (1951).
[5] De Luca, Steenbock H., *Science*, **126**, 258 (1957).

Antithyroidal Effect of Some Fresh Plants on Guinea Pigs

SEVERAL investigators have described a goitrogenic effect of various *Brassica* plants (see Greer's review[1]). Papers have appeared in recent years dealing with the goitrogenic properties of milk from cows fed on various species of *Brassica*[2-5]. As, however, no goitrogenic substances had until recently been proved in the edible parts of *Brassica* (except in rutabaga (*Brass. napus* var. *esculenta*) and turnip[1]) quite a number of problems remained unanswered. Recently, Altamura *et al.* isolated L-5-vinyl-2-thiooxazolidone also from fresh cabbage[6]. On the basis of our previous studies[5] we decided to study the effect of *Brassica* and other plants on the thyroid gland of laboratory animals with the aid of newer methods.

12 male guinea pigs were divided into three groups (4 animals per group) and maintained 48 hr. on tap-water only. Then they received vegetables (group I carrot, group II kohlrabi and group III kale). After 24 hr., in which they consumed approximately 70–

80 gm. of vegetables per animal, they were injected with 3 μc. iodine-131 in 0·5 ml. of physiological saline intraperitoneally. 4 hr. later they were killed under ether anæsthesia. Radioactivity of thyroid glands was determined after hydrolysis in 30 per cent sodium hydroxide with the aid of a scintillation counter and the thiocyanate-level in serum was determined by Aldridge's method[7].

Table 1. EFFECT OF CERTAIN PLANTS ON ACCUMULATION OF IODINE-131 ON THE THYROID GLANDS AND ON THIOCYANATE-LEVEL IN SERUM

Plant	Iodine-131 in thyroid gland (c.p.m./mgm.)	Thiocyanate in serum (mgm. per cent)
I Carrot	5,549 ± 994	0·053 ± 0·012
II Kohlrabi	3,343 ± 636	0·550 ± 0·161
III Kale	2,778 ± 601	1·605 ± 0·807

Accumulation of radioiodine after feeding kohlrabi and kale was significantly lower than after feeding carrot ($P < 0.02$). Kohlrabi and kale were used because of their different contents of thiocyanate[8]. The results indicated that thiocyanate may indeed be a cause of the antithyroidal effect of these plants, but some other agent must likewise be present. Further experiments are in progress.

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[1] Greer, M. A., *Amer. J. Clin. Nutr.*, **5**, 440 (1957).

[2] Clements, F. W., and Wishart, J. W., *Metabolism*, **5**, 623 (1956).

[3] Greene, R., Farran, H., and Glascock, R. F., *J. Endocrinol.*, **17**, 272 (1958).

[4] Wright, E., *Nature*, **181**, 1602 (1958).

[5] Langer, P., Sedlák, J., Michajlovskij, N., Štukovský, R., and Podoba, J., *Arch. Sci. Med.*, **105**, 257 (1958).

[6] Altamura, M. R., Long, J. R., and Hasselstrom, T., *J. Biol. Chem.*, **234**, 1847 (1959).

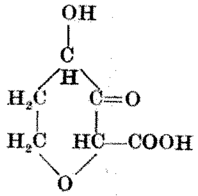
[7] Aldridge, W. N., *Analyst*, **70**, 474 (1945).

[8] Michajlovskij, N., and Langer, P., *Hoppe-Sey. Z. Phys. Chem.*, **312**, 26 (1958).

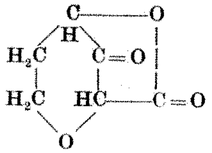
Activity of a Substance extracted from the Fermentation Products of *Acetobacter aceti* on Ehrlich's Ascites Tumour Cells

DURING the bacterial fermentation of alcohol to acetic acid, highly active molecules capable of regulating oxidation-reduction potentials must be elaborated by the bacteria, but as yet no extensive investigations in this field have been carried out. A substance possessing interesting biological activities and with some well-defined chemical characteristics has recently been isolated from vinegar[1,2]. This substance, obtained by extraction and purification processes similar to those used for cytochrome isolation, proved to have a marked oxidizing ability and, in particular, exerted an intense facilitatory activity on some catalase systems. It also markedly inhibited the growth of human tumour cells cultivated *in vitro*. When the substance is added to the medium in very low concentration in *in vitro* respiration experiments (Warburg technique) with Ehrlich's ascites tumour cells, an increase of respiration occurs: on the other hand, a most important finding is that respiration is blocked at higher concentrations. This interference with respiratory processes is much less marked on non-tumour cells. These effects would support Antoniani's concept[3] of the extreme sensitivity of tumour cells to substances capable of interfering with their respiratory systems, which, owing to discontinuous pathways, are particularly labile.

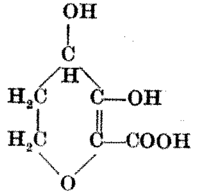
The substance, previously characterized by extraction methods and chromatographic purification, has been recently isolated in the pure state. It is a white powder with a melting point of 184° C. Its probable structural formula, derived from the elementary analytical data, from determination of the molecular weight (camphor method), infra-red spectrum, chemical groups and break-down studies, is:



It may be in equilibrium with a γ-lactone form:



or may exhibit an enolic form as follows:



The compound is easily soluble in water; the solutions are acid (about pH 3·2) and slightly yellow.

Its toxicity is very low: 2·5 gm./kgm. intravenously are not lethal for mice. The addition of the compound (concentration 1 : 100,000) to the perfusing fluid in the Straub frog heart preparation induces a digitalis-like pattern without producing heart block.

Two types of experiment have been performed with samples of the pure substance in saline adjusted to pH 7·4.

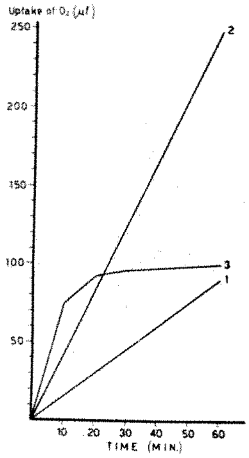


Fig. 1. 1, Ehrlich ascites cells, 1·0 ml. (2·9 mgm. nitrogen by Kjeldahl); ascites serum, 1·1 ml.; glucose solution, *M*/100, 0·5 ml.; substance, 0
2, Ehrlich ascites cells, 1·0 ml. (2·9 mgm. nitrogen by Kjeldahl); ascites serum, 1·1 ml.; glucose solution, *M*/100, 0·5 ml.; substance, 0·6 mgm.
3, Ehrlich ascites cells, 1·0 ml. (2·9 mgm. nitrogen by Kjeldahl); ascites serum, 1·1 ml.; glucose solution *M*/100, 0·5 ml.; substance, 3 mgm.

Table 1. Mice with Ehrlich's Ascites Tumour

No. of animals		Days after tumour transplantation					
		0	2	4	6	8	10
30 controls	body-weight	19·60	20·10	23·10	25·60	26·9	31·2
	cells/ml. present in ascitic fluid	25,000,000	89,600,000	146,700,000	165,800,000	—	—
30 treated (3 mgm./day of the substance)	body-weight	19·83	20·02	20·20	21·70	21·3	22·3
	cells/ml. present in ascitic fluid	25,000,000	38,450,000	27,650,000	37,900,000	—	—

The effects *in vitro* on the respiration of Ehrlich's ascites tumour cells have been studied using the Warburg technique, while in the *in vivo* experiments Ehrlich's ascites tumour in the white mice were used.

The results of the *in vitro* experiments are illustrated in Fig. 1. When the compound is added, in doses of 0·6 mgm., to the fluid containing the ascites tumour cells (equivalent to 2·9 mgm. nitrogen) and substrates, made up to a total volume of 2·8 ml., a marked increase in cellular respiration is seen in the first hour. The addition of 3 mgm. of substance, under the same experimental conditions, produces increased respiration for the first 20 min. and then a complete inhibition of respiration.

The results of the *in vivo* experiments are reported in Table 1. The neutralized solution was injected daily at a dose of 3 mgm./animal/day beginning from the second day after the intraperitoneal inoculation of the ascitic fluid containing the tumour cells.

Treated mice showed nearly complete inhibition of tumour growth (there was no increase in body-weight) and very marked decrease of number of cells per ml. of ascitic fluid.

These results confirm the marked *in vivo* activity of this new substance in inhibiting the development of the particular type of tumour cell used in these experiments.

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[1] Antoniani, C., Cernuschi, E., Lanzani, G. A., and Pecile, A., *La Clin. Ost. Ginecol.*, **60**, 225 (1958).

[2] Yamamoto, T., Lanzani, G. A., and Cernuschi, E., *La Clin. Ost. Ginecol.*, **61**, 35 (1959).

[3] Antoniani, C., *La Clin. Ost. Ginecol.*, **59**, 51 (1957).

BIOPHYSICS

Reversible Supercontraction of β-Keratose

Peacock[1] has examined the X-ray diffraction patterns from Lincoln wool supercontracted by oxidation in peracetic acid. An observation made by Whiteley[2] using Alexander's[3] peracetic acid–ammonia technique to separate the protein components in wool[4] was that, following supercontraction in peracetic acid, the fibres increase in length when the α- and γ-keratoses are dissolved out by the ammonia solution. Mercer found it possible to prepare a birefringent fibre from the protein extracted from wool by urea-reducer solutions which showed dimensional changes as the *p*H was altered[5].

We now find that β-keratose, which remains as a continuous fibre containing some 10 per cent of the original protein[3], will contract again if taken from alkaline solution into solutions of lower *p*H. Moreover, the change in length of the β-keratose fibre as the *p*H is altered is reversible, without hysteresis, and can be repeated many times with a single fibre. The experiment has been carried out with oxidized Merino and Lincoln wools, human hair and horse hair at temperatures between 30° and 60° C.

In a typical experiment a single horse hair about 2 cm. long was mounted in a setting frame and oxidized for 17 hr. at 40° C. and 12 hr. at 60° C. by an approximately 5 per cent w/v peracetic acid solution. The hair was then extracted with 0·1 *N* ammonia and contraction-extension experiments were carried out by placing the extracted fibre in solutions of various *p*H values made from mixtures of 0·01 ammonia and 0·01 *N* acetic acid. The length of the fibre was measured with a travelling microscope after equilibration at each new *p*H at 23° C. for ½ hr. The results are shown in Fig. 1.

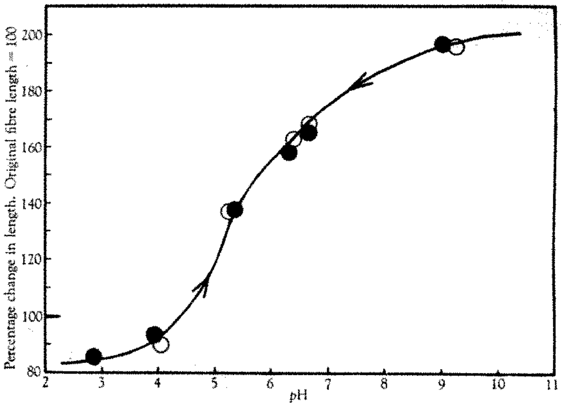

Fig. 1. Reversible length change of β-keratose from horse-hair as the *p*H is altered. Full circles : *p*H falling ; open circles : *p*H rising

When examined by X-ray diffraction, Lincoln wool appears to be exceptional in giving a pattern indicative of α-protein both after mild oxidation (see Peacock[1], for example) and after contraction of the alkali-extended fibres. On the other hand, prolonged oxidation results in the appearance of a pronounced β ring at 4·65 A., and this change in the diffraction pattern, often accompanied by some preferential orientation on the meridian (the cross-β pattern) associated with the supercontracted state, is the more usual observation in other keratin fibres oxidized with peracetic acid. After extraction of these oxidized fibres with ammonia, the insoluble residue, when alkaline, always gives only a diffuse protein-type pattern in spite of attempts to orient the material by stretching. As the *p*H is lowered, a pronounced sharpening of the 4·65 A. reflexion develops and this shows a slight preferred orientation on the meridian after carefully stretching the wetted fibres. These changes have been followed particularly in the more robust horse hair. In contrast to these findings, neither dimensional nor configurational changes have been observed in thin films of α-keratose cast from ammonia (they give a well-oriented α-pattern) and subjected to the same changes of *p*H as those applied to the extracted fibre.

The relationship between supercontraction in wool and hair and the contraction of muscle has been repeatedly pointed out by Astbury[6,7], and there is evidence that supercontraction occurs during the synæresis of actomyosin gels[8]. Moreover, the ease with which actomyosin undergoes supercontraction in the presence of hydrogen ions[9] supports the evidence of Goodall[10] that a proton-transfer mechanism might be responsible for the development of tension in muscle. The dimensional changes of β-keratose as the *p*H is altered suggest again at least a partial contraction to the cross-β configuration, though apparently in this case, since the X-ray diagram given by the extended fibre is so diffuse, not from a well-defined α-configuration but from some intermediate less-regular state.

We wish to thank Prof. W. T. Astbury for his advice and encouragement in this investigation, which is supported in part by the Muscular Dystrophy Associations of America, Inc., and will be reported in detail elsewhere.

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[1] Peacock, N., M.Sc. thesis, University of Leeds (1952).
[2] Whiteley, K. J., Ph.D. thesis, University of Leeds (1959).
[3] Alexander, P., and Hudson, R. F., "Wool—its Chemistry and Physics", 265 (Chapman and Hall, London, 1954).
[4] Corfield, M. C., Robson, A., and Skinner, B., *Biochem. J.*, **68**, 348 (1958).
[5] Mercer, E. H., *Textile Res. J.*, **24**, 835 (1954).
[6] Astbury, W. T., *Proc. Roy. Soc.*, B, **134**, 303 (1947) (Croonian Lecture, 1945).
[7] Astbury, W. T., Proc. Intern. Wool Textile Res. Conf., Australia, B, 220 (1955).
[8] Pautard, F. G. E., *Nature*, **182**, 788 (1958).
[9] Pautard, F. G. E., *Nature*, **183**, 1391 (1959).
[10] Goodall, M. C., *Nature*, **182**, 677 (1958).

PLANT PHYSIOLOGY

Effect of Infection by *Xanthomonas malvacearum* (E.F.S.) Dowson on Respiration-rate of Cotton Leaves

Potted plants of susceptible Domains Sakel cotton (*Gossypium barbadense*, L.) and its synthesized blackarm-resistant strains containing B_2 or $B_2 + B_3 + B_6$ genes were sprayed when about three months old with a standardized water-suspension of *Xanthomonas malvacearum* taken from three-day-old cultures of a recent isolate. The plants were kept in the shade under glass bell-jars for 48 hr. after inoculation. The leaves used for determining respiration-rates were chosen from among fully expanded leaves 5 cm. in diameter situated at the ninth to thirteenth nodes of the stem.

The number of spots on inoculated leaves ranged from 80 and 200 per leaf. A Thoday respirometer was used for determining oxygen uptake, which was taken as a measure of respiration-rate. Two leaves, of about 1 gm. fresh weight, were placed in each respiratory chamber with the petioles dipped in distilled water in a specimen tube. The respiratory chamber also contained 10 ml. of 5 per cent potassium hydroxide and a piece of Whatman folded filter paper No. 40 to increase the absorption surface. The chambers were placed in a temperature-controlled water-bath adjusted at 30° C. Readings were taken 2 hr. after the experiment had been started and continued thereafter at 8-hr. intervals. The first reading in all experiments was discarded to avoid misinterpretation of handling effects.

In 1957, two sets of experiments were carried out using leaves of susceptible Domains Sakel. In the first set, uninoculated (control) leaves as well as leaves 4 and 10 days after inoculation were used. The second set included control leaves, and leaves taken 10 and 16 days after inoculation. The experiments were run for 2 days. The respiration-rate of the leaves of resistant strains of Domains Sakel containing B_2 and $B_2 + B_3 + B_6$ was also examined in uninoculated leaves (control) and in leaves 10 and 16 days after inoculation. These experiments were run for 4 days.

In 1958, experiments were carried out with the susceptible Domains Sakel using uninoculated leaves (control) and leaves 4, 8, 12 and 16 days after inoculation. The leaves were cut at noon, washed thoroughly with distilled water, blotted dry with filter paper, weighed and placed in the respiratory chamber. The experiments were run for 4 days and thus respiration was actually measured continuously from the fifth to the twentieth day after inoculation.

Angular leaf spots first appeared on susceptible Domains Sakel 5–7 days after inoculation as minute, water-soaked, dark green spots mainly on the undersurface of the leaf. The spots increased in size, becoming 2–4 mm. in diameter in a few days, but did not attain their characteristic brown colour until 15–16 days after inoculation. On the resistant strains of Sakel, development of disease symptoms could only be ascertained between the eighth and tenth day after inoculation, when minute, pin-head, dark green, dry round spots appeared. The spots turned dark brown with age but remained very small.

The results of experiments in 1957 (Table 1) showed that there was a general depression in the respiration-rate of the inoculated leaves of susceptible Domains Sakel and its resistant strains compared with the controls. The depression seemed to reach its maximum in susceptible Sakel earlier than in the resistant strains.

The picture was clearer in the 1958 experiments, where the respiration-rate of Domains Sakel leaves was followed up from the fifth to the twentieth day after inoculation. Fig. 1 shows that maximum depression in respiration-rate was reached between the fifth and seventh days after inoculation. Respiration-rate then increased continually until the twentieth day, when no further readings were recorded.

Fluctuations in respiration-rate seem to coincide with the development of disease symptoms. Thus the early depression in respiration coincides with the period in which the bacteria enter the stomata and multiply in the stomatal chambers, blocking the stomata and suppressing gas exchange. This phase of the course of disease takes place in both susceptible and resistant strains of Domains Sakel. About the time disease symptoms first appear as pinhead spots, respiration reaches its minimum (that is, 5–7 days

Table 1. Respiration-Rate of Leaves of Resistant and Susceptible Strains of Domains Sakel Cotton inoculated with *X. malvacearum* (1957 Experiments)

Cottons	Oxygen uptake (ml. dry gas/ 10 gm. fresh wt./hr.)			
	Days after inoculation			
	Control	4	10	16
Susceptible Domains Sakel				
Means of three experiments	6·15	5·28	5·90	—
Difference from control (per cent)		−14·2	−4·1	—
Means of three experiments	5·71	—	5·36	7·88
Difference from control (per cent)			−5·1	+38·00
Sakel containing B_2				
Means of two experiments	5·39	—	4·79	4·88
Difference from control (per cent)		—	−11·1	−9·5
Sakel containing $B_2+B_3+B_4$				
Means of two experiments	5·39	—	4·62	5·53
Difference from control (per cent)		—	−14·3	+2·6

after inoculation in the case of susceptible Sakel and later in that of resistant strains). The increase in respiration-rate of the leaves of susceptible Domains Sakel that follows coincides with the increase in the size of the spots and the disintegration and collapse of the spot cells. The increase may be attributed partly to the evolution of the gas previously accumulated in the tissues, and partly to increased respiratory activity of the cells resulting from infection. The amount of gas contributed by the bacteria themselves to carbon dioxide output of affected leaves is difficult to assess but is unlikely to be of much significance.

In the resistant strains, the spots remain pinhead in size and the respiration-rate increases to about normal after their appearance. In this instance the depression in carbon dioxide output is less than in susceptible Sakel, but continues for a longer period before it returns to more or less the normal rate.

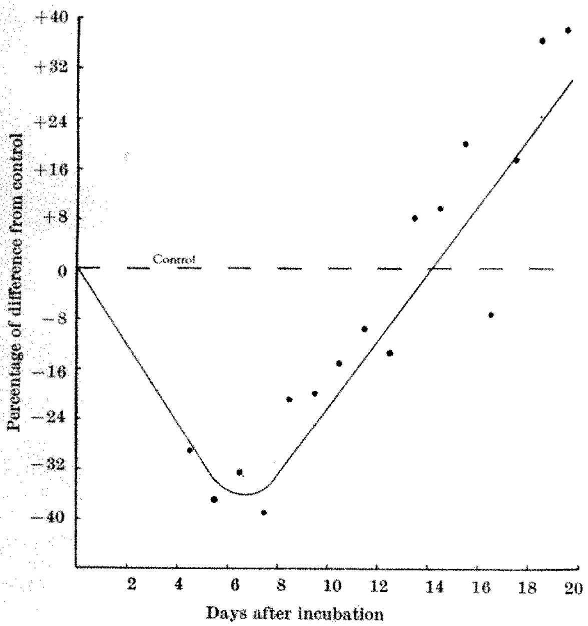

Fig. 1 Relationship between respiration-rate and age of inoculation

The damage caused to the host is little, if any, and does not provoke marked cell respiration.

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Chemical Nature of the Cell Wall of the Green Algae, *Codium*, *Acetabularia* and *Halicoryne*

The chemical nature of the carbohydrates which constitute the main cell wall of the algae belonging to Codiaceae and Dasycladaceae has remained thus far largely obscure. Though the cell wall of *Codium* was said to consist of callose and pectin together with some cellulose[1], no chemical characterization of callose and algal pectin has yet been achieved, their occurrence being recognized merely by staining reactions and solubility tests. Likewise, no unequivocal proof for the presence in *Codium* of true cellulose has hitherto been obtained, although its occurrence has occasionally been reported[2,3]. As for the cell-wall constituents of the members of Dasycladaceae almost nothing is known at present except the incrustation of calcium in the cell wall.

As a continuation of the biochemical researches of algal cell wall by one of us[4], we have been engaged in the study of the cell-wall constituents of the green algae from different taxonomic groups with the view of obtaining information about the phylogeny and systematic position.

The present communication deals with the studies on *Codium fragile*, *Acetabularia calyculus* and *Halicoryne wrightii*.

The air-dried sample of each seaweed was soaked in 1 per cent hydrochloric acid for 24 hr. at room temperature to remove calcium carbonate. It was then treated successively with 1·25 per cent sodium hydroxide and 1·25 per cent sulphuric acid at 100° for 30 min. and the residue was bleached with 1 per cent sodium chlorite. The crude fibre thus obtained retained the membrane structure almost unchanged and showed neither anisotropy nor staining reactions for cellulose.

Hydrolysis of the crude fibre with sulphuric acid and examination of the products by paper chromatography revealed the main constituents to be mannose. A trace of glucose as detected in the hydrolysate arose presumably from starch which could not be removed by the above treatment. The crude fibre is insoluble in sodium hydroxide at any concentrations, but easily soluble in 50 per cent zinc chloride, less in 50 per cent stannic chloride and slightly in Schweizer's reagent. Hence the purification was effected by dissolution in zinc chloride followed by precipitation with acetone. As the product contained about 2 per cent of ash, of which the major part was silica, it was removed by the method of Feigl and Krumholz[5]. The purified substance is a white powder with an ash content less than 0·1 per cent. Elementary composition corresponded to hexosan. Calculated for $C_6H_{10}O_5$, C 44·44, H 6·22 ; found, C 44·23, H 6·09 (*Codium*) ; C 44·24, H 6·17 (*Acetabularia*) ; C 44·31, H 6·12 (*Halicoryne*). $[\alpha]_D$ in 50 per cent zinc chloride −46·1° (*c.* 1·41, *Codium*) ; −50·6° (*c.* 1·54, *Acetabularia*) ; −46·1° (*c.* 1·41, *Halicoryne*). Hydrolysis with 40 per cent hydrochloric acid at 0° yielded 91–93 per cent of mannose. On the paper chromatogram no other spots than that of mannose could be detected. Mannose phenylhydrazone (m.p. 199°) was obtained in nearly the theoretical yield[6]. When oxidized with periodate 0·96 mole of it was consumed and 0·125 mole of formic acid was produced per unit $(C_6H_{10}O_5)$ of the preparation from *Codium*. Assuming a straight-chain structure with 1,4 linkage the yield of formic acid indicates a degree of polymerization of 16.

These results, taken together with the resemblance in specific rotation to the mannan of ivory nut (− 44·1°) and salep (− 35·0°), both of which are known to be β-1,4-linked, would indicate that the mannans from *Codium*, *Acetabularia* and *Halicoryne* are constructed in a similar manner.

Other *Codium* species such as *C. latum* and *C. tomentosum* also contain mannan as main cell-wall constituent, whereas in *Udotea*, *Chlorodesmis* and *Halimeda*, all of which are usually included in Codiaceae, mannan is replaced by xylan with β-1,3-linkage.

The significance of these findings in connexion with phylogeny and taxonomy will be discussed elsewhere.

We wish to express our gratitude to Profs. S. Inoh and S. Segawa as well as to Drs. M. Chihara and S. Kamura for their generous support in this work.

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[1] Prinz, H., in Engler-Gilgs "Die natürlichen Pflanzenfamilien", **3**, 2, 309 (1927).

[2] Augier, J., and du Mérac, M. L. R., *Bull. Lab. Maritime Dinard*, **40**, 25 (1954); *C.A.*, **51**, 1395, (1957).

[3] Hayashi, K., Nagata, Y., and Yamada, O., *Res. Bull. Fac. Agric. Gifu Univ.*, **5**, 90 (1955).

[4] Miwa, T., *Jap. J. Bot.*, **11**, 41 (1940).

[5] Feigl, F., and Krumholz, P., *Mikrochemie*, **7**, 82 (1929).

[6] Bourquelot, E., and Hérissey, H., *C.R. Acad. Sci., Paris*, **129**, 339 (1899).

BIOLOGY

Attrition of Incisors of Grazing Sheep

UNDER New Zealand conditions of sheep-farming, incisors of grazing sheep wear much more rapidly on improved pasture, chiefly ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*), than on the finer native pastures of low carrying capacity. The cause is not nutritional in the generally accepted sense, however[1]. It would rather seem that certain substances in the herbage of 'improved' pastures dissolve the teeth, the process being aided by the abrasive action of the plant fibre. Wear is also increased in mouths with certain undesirable anatomical characteristics which appear to be hereditary in origin.

Differences in chemical compositions of enamels and dentines of sheep's teeth of various breeds and from farms on country of dissimilar soil types are not correlated with the extent of tooth wear[2].

The thin layer of enamel on the incisal surface of sheep's teeth is worn through approximately two months after eruption, hence the problem is concerned with the rate of wear of primary dentine and, at a later stage, secondary dentine.

Electron micrographs of the biting edge of worn incisors showed that the dentine surface was subjected to both chemical and abrasive action. The chemical action was thought to be due to the organic compounds in the herbage complexing the calcium phosphate (apatite), but before this could be checked by studies with the electron microscope it was essential to understand the microstructure of the organic and inorganic components of dentine more fully[3] and to develop a technique which enabled the intensity and nature of etching to be recorded (Hall, D. M., unpublished work). Experiments were then made to compare the etching produced by fractions of juices of pasture plants on surfaces of fresh dentine polished smooth by metallographic techniques. The fractions examined were: (1) organic acids from herbage at their original concentrations and *p*H (6·0–6·5) for chelating action on the calcium; (2) cell fluids, obtained by ether cytolysis, containing in addition sugars, inorganic and organic phosphates, amino-acids and polypeptides, capable of chelating or dissolving calcium; (3) freshly expressed juices of grasses and clovers containing enzymes in addition to substances of (1) and (2).

These studies have shown that fraction (3) produced the strongest etching, and indicated that this reaches a maximum during the lush spring growth. It was also evident that the central non-calcified core of the odontoblast process was etched by fresh grass juice at a faster rate than the dentinal matrix and thereby released long lengths of the membrane-like sheaths enclosing the process.

The results indicate that the majority of the etching is due to proteolysis and that such weakening of the bonding material of the tooth would facilitate its wear by attrition. Rouiller has observed that an organic component bonding the particles of dentine is digested by plant proteinases such as papain[4,5]. It is considered, therefore, that the digestive action of the proteinases of actively metabolizing leaves on the organic components of dentine could be an important factor contributing to the wear in teeth of grazing sheep.

This investigation is being continued.

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[1] Barnicoat, C. R., *N.Z. J. Sci. Tech. Sec.*, A, **38**, 583 (1957).

[2] Barnicoat, C. R., *N.Z. J. Sci. Tech. Sec.* (in the press).

[3] Hall, D. M., *J. Dental Res.*, **38**, 386 (1959).

[4] Rouiller, C., Huber, L., and Rutishauser, E., *Acta Anat.*, **16**, 16 (1952).

[5] Rouiller, C., "The Biochemistry and Physiology of Bone", ed. Bourne, G. H., 136 (Academic Press, Inc., New York, 1956).

Rhizosphere Effect on Soil Algae

VERY few studies of the effect of the rhizosphere on soil algae have been made in the past[1], and in a recent review Starkey[2] states that it is possible that algal populations may even be decreased in the root zone. This communication presents evidence for a well-marked rhizosphere effect on soil algae by the roots of cultivated tea plants nominally referable to the Assam variety of *Camellia sinensis* (L) O. Kuntze.

Roots from one-year old seedlings were removed from the nursery beds and the soil from the top 5 cm. of the roots was scraped off with a sterile spatula. The top 5 cm. of soil from adjacent bare areas of the same bed was used as the control. Modifying the method described by Agnihothrudu[3], 5 gm. each of

Table 1. Presence or Absence of Algae in Various Dilutions of Bristol's Solution

	Dilution						
	1/10	1/100	1/1,000	1/10,000	1/100,000	1/1,000,000	1/10,000,000
Rhizosphere soil	+	+	+	+	+	+	+
Control soil	+	+	+	+	+	−	−

control and rhizosphere soil samples were used for preparing a series of ten dilutions in Bristol's solution[4], the initial dilution being 1/10, and each higher dilution made by taking 10 ml. of the lower dilution and adding it to 90 ml. of sterile Bristol's solution. Three replications of each dilution were made and growth was estimated after three months by the presence or absence of algae in the flasks (Table 1). Previous observations on the soil algae of our subtropical soils showed that this period was sufficient for growth to occur and also that Bristol's solution was the most suitable of several media tried.

There is at present no reliable method of estimating numbers of soil algae in a mixture of unicellular, colonial and filamentous forms and the presence or absence of visible growth remains the only satisfactory method for this type of study.

These results give evidence of an effect of the rhizosphere on soil algae in the case of tea roots. The experiments have been repeated with other crop plants, both monocotyledons and dicotyledons, and have confirmed the presence of this effect. The results will be published in greater detail elsewhere.

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[1] Katznelson, H., *Soil. Sci.*, **62**, 343 (1946).
[2] Starkey, R. L., *Bact. Rev.*, **22**, 154 (1958).
[3] Agnihothrudu, V., *Proc. India Acad. Sci.*, **378**, 1 (1953).
[4] Bristol, B. M., *Ann. Bot.*, **34**, 35 (1920).

The Field-Slug *Agriolimax reticulatus* as a Vector of the Lungworm *Cystocaulus ocreatus*

Gerichter[1] demonstrated that the land snail *Monacha syriaca* can act as a vector for the sheep lungworm *Cystocaulus ocreatus*. Development of the lungworm larvæ can take place in other species of land molluscs and also in some freshwater species[1,2]. The importance of most of these molluscs as vectors under field conditions does not, however, appear to have been investigated.

C. ocreatus was recorded only recently in British sheep[3]. Afterwards a study of the transmission of infection under natural conditions was commenced at Weybridge. Lungworm-free lambs were grazed with infected ewes and, at frequent intervals, fæcal samples from the lambs were examined for first-stage lungworm larvæ. After several weeks, larvæ were recovered from the fæces, and on autopsy of one lamb, adult worms were found in the lungs. Samples of the land molluscs inhabiting the pasture grazed by the sheep were collected and examined for lungworm larvæ. The only species infected was the field-slug *Agriolimax reticulatus*, so that it appeared that infection of the lambs had resulted from the ingestion of field-slugs. To verify that this was so, the feet from artificially infected slugs were fed to two stalled lambs reared free of lungworms. After several weeks, first-stage larvæ of *C. ocreatus* were recovered from both lambs.

A. reticulatus was by far the most numerous of the mollusc species collected from the Weybridge pasture, specimens being found throughout the year. According to Ellis[4], the field-slug is the commonest and most ubiquitous of the land molluscs found in Britain, inhabiting every possible situation. As *A. reticulatus* is almost certain to be found wherever sheep can graze, it is most likely that it is of considerable importance as a vector of *C. ocreatus* in Britain.

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[1] Gerichter, Ch. B., *Parasitology*, **41**, 166 (1951).
[2] Kassai, T., *Z. Parasitenk.*, **18**, 5 (1957).
[3] Rose, J. H., *Nature*, **179**, 925 (1957).
[4] Ellis, A. E. E., "British Snails", 256 (Oxf. Univ. Press, 1926).

The Pituitary of *Myxine glutinosa*

The Myxinoids are possibly the most primitive living vertebrates, and the pituitary of *Myxine glutinosa* has hitherto[1-3] been described as having a separate infundibulum and adenohypophysis, a condition which has been regarded as primitive or degenerate. Furthermore, the adenohypophysis which lies embedded as groups of cells in connective tissue, has been described as being composed solely of chromophobes, and having no cytological or histological differentiation. Recent observations, however, have shown that the connective tissue septum does not separate completely the two parts of the pituitary but that the posterior end of the infundibulum comes into intimate contact with the adenohypophysis (Fig. 1). Also, the adenohypophysis is cytologically differentiated.

An examination of the infundibulum using Bodian's stain indicated that nerve fibres lie between and beneath the ependymal cells and posteriorly are closely apposed to adenohypophysial cells. Using several methods to demonstrate neurosecretory material this was shown to be present in the infundibular canal particularly where it narrows posteriorly.

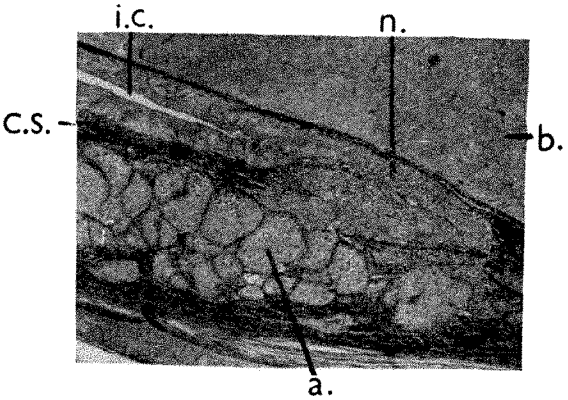

Fig. 1. Longitudinal section of the caudal end of the pituitary of *Myxine glutinosa* showing apposition of neurohypophysis and adenohypophysis. *a*, Adenohypophysis; *b*, brain; *c.s.*, connective tissue septum; *i.c.*, infundibular canal; *n*, neurohypophysis

Neurosecretion was also present in the region of the neurones lying outside the infundibulum. In the caudal part of what now must be regarded as a neurohypophysis a few cells identical with the ependymal cells are found separate from the lining of the ventricle. No neurosecretory material was found in the adenohypophysial region.

The adenohypophysis when stained with Heidenhain's azan stain or Mallory's triple stain was shown to contain at least two different types of basophils in addition to chromophobes and large vacuolated cells.

Furthermore, basophilic and periodic acid–Schiff positive colloidal aggregations occur in some adenohypophysial follicles as do epithelium-lined cavities. No distinct acidophils were observed. After applying the periodic acid–Schiff it was shown that the basophils contained periodic acid–Schiff positive material as in higher vertebrates. Some cells also stained after applying the Gomori aldehyde–fuchsin technique. It seems highly probable then that the pituitary of *Myxine* secretes carbohydrate-containing hormones in a similar manner to other vertebrates.

These observations, which will be reported in full elsewhere, indicate that the pituitary of *Myxine glutinosa* is possibly the simplest form of vertebrate pituitary and is a form which is not degenerate but is one in which the adenohypophysis has not become histologically differentiated.

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[1] De Beer, G. R., "The Comparative Anatomy, Histology and Development of the Pituitary Body" (Oliver and Boyd, London, 1926).

[2] Grasse, P. P., "Traité de Zoologie", 13 (Masson, Paris, 1958).

[3] Stendall, W., *Arch. mikrosk. Anat.*, **82**, 289 (1913).

Swimming in Spider Crabs of the Genus *Macropodia*

The only adult Brachyura which have previously been recorded swimming are the Portunidae, which do so by rapid paddling of the last pair of peræopods. These are adapted for swimming by having the distal segments flattened, with the edges densely fringed with hairs.

Swimming has now also been observed in *Macropodia rostrata* (L.) and *Macropodia longirostris* (Fabricius), members of the Maiidae (spider crabs). Both these species occur sublittorally in the Isle of Man. The genus has very long legs which bear a number of long hairs, and swimming involves the four pairs of walking legs and the chelæ. All the limbs beat at the same rate, but not in phase, and are kept continually moving. At the start of the stroke they are raised in the flexed position, and are extended for the more rapid downbeat. Swimming may reach a speed of 5 cm. per sec. in a specimen of *Macropodia rostrata* with a carapace-length of 13 mm.

The pattern of movement of the legs has been followed, and it is the same in both species. The chelæ and walking legs will be referred to as *c*, 1, 2, 3, 4 respectively, on the right (R) and left (L) sides.

The swimming limbs can be divided into three groups: (1) L^2 and R^2 beat in phase with each other, but out of phase with all the other legs; that is, when group 1 is beating down, groups 2 and 3 are on the up-beat. (2) R^c, R^1, L^3 and L^4 beat as a unit, with L^4 slightly after the rest. (3) L^c, L^1, R^3 and R^4 also beat as a unit, with R^4 just behind the others. One of groups 2 and 3 starts to beat slightly before the other, and either may take the lead during a period of swimming. The rate of beating varies from once to twice per sec. The body of the crab is horizontal during swimming, and the sequence of movement of the legs prevents rolling or pitching. This is because the legs beating down at any moment are symmetrically arranged around the centre of gravity of the crab (for example, L^2 opposite R^2, and R^c and R^1 opposite L^3 and L^4).

In the laboratory, swimming may occur apparently spontaneously, as a result of attack by another crab, or on repeated disturbance. It occurs more readily in smaller specimens, and large ones can seldom be induced to swim.

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A New Cercaria of the Subfamily Gymnophallinae (Trematoda: Digenea) developing in a unique 'Parthenita' in *Littorina saxatilis* (Olivi)

A 'parthenita' containing furcocerous and tailless stages of a gymnophalline cercaria was found in the hæmolymph spaces of the digestive gland, gonad and gonoduct of the gastropod *Littorina saxatilis*. The unique feature of this 'parthenita' is that it has the same essential structure as the cercariæ which it produces. It is not yet possible to decide whether this 'parthenita' is, in fact, a cercaria possessing the kind of multiplication more usually associated with a redia or sporocyst; or whether it is a sporocyst or redia with a structure similar to a cercaria. Until this is decided the word 'parthenita' will be used here to indicate an analogy with the more usual redia or sporocyst found in digenetic trematodes.

This is the first record of a gymnophalline cercaria developing in a gastropod mollusc; previous records having been recorded from lamellibranchs. A detailed account of the information available to date on this species is being prepared for publication.

The 'parthenita' was found in 490 of 14,000 specimens of *Littorina saxatilis* examined between September 1958 and September 1959 at Aberystwyth and in 59 of 1,100 snails in August 1959 at Plymouth. Infection was almost exclusively confined to small molluscs, measuring from 0·6 to 5·0 mm. in length, occurring in the supralittoral zone; in favourable circumstances up to 40 per cent infection exists.

The fully formed 'parthenita' (Fig. 1) is spherical in shape, measuring 0·2–1·2 mm. in diameter and containing 20–2,000 tailless cercariæ. 5–70 fully formed 'parthenitæ' may occur in any one host. The whole surface of the 'parthenita' is covered with alternating rows of large backwardly directed spines. An oral sucker is present and specimens measuring less than 0·2 mm. in diameter have, also, a ventral sucker. The bifurcated alimentary canal consists of pharynx, œsophagus and two dilated cæca which are connected to the body-wall of the 'parthenita' by strands of tissue. There is no birth pore. The excretory system is best seen in specimens less than 0·3 mm. in diameter; in larger specimens the arrangement becomes difficult to observe. The translucent excretory vesicle is Y-shaped, the arms of the Y extending forwards as far as the posterior border of

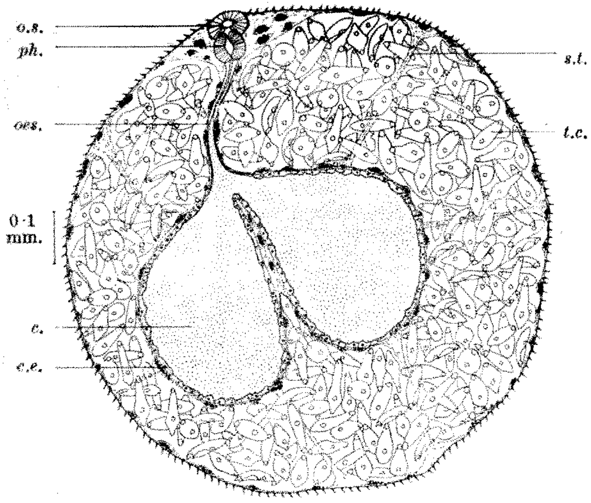

Fig. 1. A fully formed 'parthenita' containing tailless gymnophalline cercariæ from *Littorina saxatilis*. *c*, Cæcum ; *c.e.*, ciliated epithelial cells of cæcum ; *oes.*, œsophagus ; *o.s.*, oral sucker ; *ph.*, pharynx ; *s.t.*, tissue strand supporting cæcum ; *t.c.*, tailless cercaria

the pharynx, curving inwards as they do so. The flame cell formula is 2((2 + 2) + (2)) = 12. The 'parthenita', therefore, closely resembles a tailless gymnophalline cercaria.

Many 'parthenitæ' have been examined, and from these it has been possible to construct a series in development terminating in the formation of the fully formed 'parthenita' containing tailless cercariæ.

The earliest stage so far discovered is the mother 'parthenita', several generations of which may occur. The mother 'parthenita' measures 0·16–0·4 mm. in diameter and has all the anatomical features of the fully formed 'parthenita' except that it contains 5–12 developing 'parthenitæ' instead of cercariæ.

The contained 'parthenitæ' undergo a most unusual development, passing through a furcocercous stage similar in size and structure to the furcocercous cercariæ, which later develop into tailless cercariæ in the fully formed 'parthenita'. Unlike the furcocercous cercariæ, however, the tails are spineless and immobile ; these furcocercous stages cannot swim when released artificially. During the course of further development the tail of each furcocercous stage degenerates and a cavity containing germinal balls is formed in the body. This results in the formation of a number of contractile tailless 'parthenitæ' with the characteristic features of a tailless gymnophalline cercaria but possessing, also, a cavity filled with germinal balls. These 'parthenitæ' increase in size and the pressure so produced causes the wall of the mother 'parthenita' to burst, releasing all the 'parthenitæ', which are by this time at a similar stage in development. The body of the liberated 'parthenita' measures 0·12–0·4 mm. in length, depending on the state of contraction. It is extremely mobile ; moving through the hæmolymph spaces of the host by alternate attachment of oral and ventral suckers. The 'parthenitæ' gradually develop into immobile spheres measuring up to 1·2 mm. in diameter ; the ventral sucker degenerates in specimens more than 0·2 mm. in diameter. This is the stage already described as the fully formed 'parthenita' containing the tailless cercariæ.

The development of the cercaria within the 'parthenita' shows a fundamental similarity to the development of the 'parthenita' itself. On release from the mother 'parthenita' the 'parthenita' contains a number of germinal balls. These first develop into furcocercous cercariæ with opaque mobile bodies measuring 0·12 mm. in length ; the tail stem measures 0·042 mm. in length and the furcæ 0·043 mm. Both the body and the tail are covered with alternating rows of small spines. No eyespots or penetration glands are present. The excretory vesicle is Y-shaped and the flame cell formula is 2((1) + (1)) = 4. If released artificially the furcocerous cercaria swims by lashing its tail but it survives only for a short time in these circumstances. While still inside the 'parthenita' the tails degenerate and extremely contractile transparent tailless cercariæ result. The following characters of these tailless cercariæ demonstrate how they differ from all previously described gymnophalline tailless cercariæ and metacercariæ ; they indicate, also, how closely the tailless cercariæ resemble the 'parthenita' which produces them. They are extremely small, the body measuring 0·05–0·14 mm. in length according to the degree of contraction. Alternating rows of spines cover the whole body except for a region on the ventral surface between the oral and ventral suckers. The suckers are subequal. Three pairs of penetration glands are present, the posterior border of the largest reaching as far as the bifurcation of the gut. The arms of the translucent Y-shaped excretory vesicle are unbranched and extend forward as far as the posterior border of the pharynx, converging on it as they do so. The flame cell formula is 2((2 + 2) + (2)) = 12. Rudiments of the genitalia lie behind the ventral sucker.

The formation of the tailless cercariæ within the fully formed 'parthenita' probably terminates the cycle of development in the molluscan host, the tailless stage being infective to the definitive host.

All previously recorded gymnophalline cercariæ develop in sporocysts in lamellibranchs. The metacercariæ, also, occur in lamellibranchs, with two exceptions which have been recorded from gastropods. Since the 'parthenita' and cercaria described here are almost identical in structure and development it is possible that this is a case in which the type of multiplication that usually occurs in the sporocyst has extended into the cercaria. In other words the germinal cells in the cercaria, instead of giving rise to the reproductive organs of the adult, remain germinal and continue to multiply by polyembryony with the result that the cercaria in effect becomes a parthenita. If this life-cycle includes, also, a more 'normal' sporocyst not yet found and preceding the above described 'parthenita' then this may be the case, the life-cycle then being a modification of that normally found in the Gymnophallinae. If, on the other hand, no such sporocyst or its equivalent is present then the life-cycle might be more primitive than that of any other known digenean, the 'parthenita' indicating how multiplication in the molluscan host could have developed from an adult. Speculations, however, are unwise until the entire life-cycle of this form is known. Attempts at solving the life-cycle experimentally are in progress.

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Forecasting the Annual Yield in Sea Fisheries

It is well known that the annual yield of the sea fisheries varies very much from year to year. An example of such variation is shown in Fig. 1 in which the broken curve shows the number (in millions) of skrei—spawning cod (*Gadus callarias*)—caught in the Lofoten fishing area of northern Norway in the years 1885–1958. It is obvious that such fluctuations make the planning of the fishing industry very difficult and that reliable forecasts would be helpful, even if they are only rough approximations.

In an earlier paper[1] I made the suggestion that it might be possible to compute yield forecasts by means of an aggregate of sine functions, using the periodic or period-like components found in series of measurements of the width of the annual growth zones in forest trees. The underlying idea was that the width of the growth zones and the size (in number) of the year-classes of fish populations might be determined by common factors. A rough outline of a theory was given.

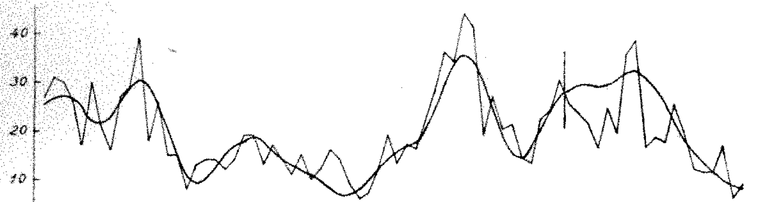

Fig. 1

Hjort[2] succeeded in showing that there are great fluctuations in the size of the year-classes in fish populations and concluded that these fluctuations were the main cause of the fluctuations in the yield. But fluctuations in the yield are also caused by other factors, such as weather conditions and altered fishing technique. Therefore, a successful forecast of the relative size of the stock of fish may not be a successful forecast of the yield; but it might show an approximately correct trend.

In 1939 I became acquainted with the results of the analyses of some series of measurements of the width of the growth zones of the Scots pine (*Pinus silvestris* L.) found by Ording[3]. His samples were taken in a forest in the neighbourhood of the Lofoten fishing area. Then, in order to verify my idea I fitted the series of annual yields of the skrei fisheries in the years 1885–1939 by a function constructed by means of four components (substituted by sine functions) found by Ording. The lengths of these components were 11, 17·5, 23 and 57 years. Also the phases of the components were taken from the results obtained by Ording. The actual yields were used merely for the computations by the least-square method of the weights of the components. The result, as well as the method of computation, is described in ref. 1. Later on I also included the 8·5 years component. The result so obtained is given by the smooth curve for the years 1885–1939 in Fig. 1.

Since this curve is constructed by means of an aggregate of sine functions, it can be extended to future years and might be used as a forecast of the annual yields. Actually, it is intended to be a forecast of the relative size of the stock of skrei. The thin curve 1940–58 shows the actual yields and the thick curve the 1939 forecasts for the same years.

It is evident that differences of the same magnitude as those observed in the years 1885–1939 should be expected. The largest differences between actual yield and forecast are found for the war years 1941–45 and in the years 1948–49. In the fishery reports from the Lofoten area I have found nothing which shows conclusively that the fishing was hampered by the war. But, bearing in mind the changes in the general conditions brought about by the war, I think we can assume that this was in fact the case. In 1948 and 1949 the weather conditions were uncommonly unfavourable. Bearing this in mind, the following conclusions seem to be justified: (1) That the width of the growth-zones of the pine and the size of the year-classes of the skrei are determined by the same main factors. (2) That it is possible to compute reliable forecasts of the yield of fisheries by means of the components found in series of measurements of the width of the growth-zones of the pine.

There is no doubt that the technique of forecasting can be improved. But in order to make improvements possible, it is necessary to sample new series of measurements of the width of growth-zones in forest trees and try to get better estimates of the lengths and the phases of the more important components.

The same method might prove to give satisfactory forecasts in other fisheries, too. However, the fishing technique used in the Lofoten area is very conservative, so that it can be assumed that the yield is a fairly constant percentage of the stock of fish. In other fisheries this may not be so. It is obvious that, for example, the introduction of a new technique may cause disturbances which are difficult to overcome.

I am very much indebted to Gunnar Rollefsen, director of the Norwegian Sea Fishery Investigations, for supplying me with data.

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[1] Ottestad, Per, Rep. Norw. Fish. and Marine Invest., **7**, No. 5 (1942).

[2] Hjort, Johan, *Conc. Perm. Internat. l'Explor. Mer, Rapp. et Proc.-Verb.*, **20** (1914).

[3] Ording, Asbjørn, *Medd. norske skogforsøksvesen*, B, **7**, H.2 (1941).

ANATOMY

Fine Innervation of the Carotid Body of the Rhesus Monkey

Although first mentioned in 1744[1], the first clear and accurate description of the carotid body was not published until 1862[2]. However, modern knowledge of the structure of this organ dates from 1926[3]. Since that time detailed macroscopic and microscopic studies of the structures found in the carotid bifurcation have been made on many different animal species. But in the Anthropoidea, except for the work of Sato[4], only macroscopic studies of this region have so far been undertaken (1852[5], 1899[6], 1928[7], 1932[8], 1935[9,10]). Sato's work is concerned only with the general histology of the organ and not its detailed innervation.

This is a preliminary report of part of an investigation into the histology and cytology of the carotid body and carotid sinus of *Macaccus rhesus*.

Rhesus monkeys were anæsthetized and then perfused through the heart first with warm normal saline to wash out the blood, and then either with 10 per cent formal–saline or Bouin's fixative.

The carotid bifurcation and adjacent tissues and structures were carefully dissected out and either treated : (1) according to the Bielschowsky method for pieces with pyridine treatment[11], or (2) 7μ serial wax sections were subjected to Bodian's silver protargol method using silver proteinate obtained from Roque's Laboratory, Paris[12].

With both techniques, the only glomus cells found to be innervated were the chief cells (De Kock's Type 1 cells[13]). Many fine nerve fibres wind round the large individual chief cells, or groups of chief cells, either in a regular manner (Fig. 1) or to form a tangled meshwork (Fig. 2). The fine nerve fibres appear to be in direct contact with the chief cells with no interposed interstitial cells[13–15]. No evidence was found of De Castro's "menisque terminaux"[16] within the cytoplasm of the cells.

The fine nerve terminals are of the order of 0·2–0·5μ in size. They fuse with each other to form larger fibres (Fig. 3) which run between the glomeruli of chief cells, and, joining other large fibres, emerge from the carotid body to form its contribution to the sinus nerve.

No evidence was found to support the observation[17] that the heavily medullated fibres of the sinus nerve, which most authorities consider as innervating both carotid body and sinus, do not take part in the innervation of the carotid body but are merely passing through on their way to the carotid sinus.

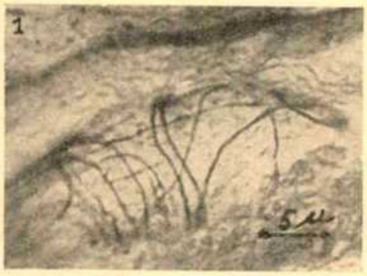

Fig. 1. A group of glomus cells showing the fairly regular arrangement of their innervating nerve fibres

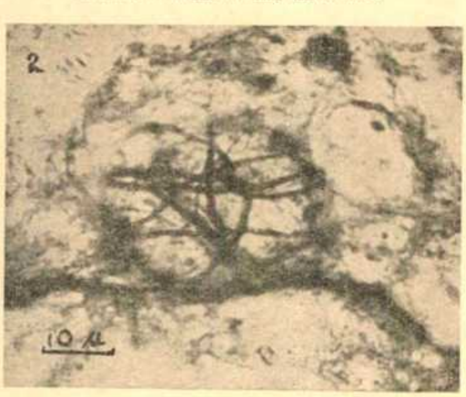

Fig. 2. Glomus cells enclosed in a tangled meshwork of nerve fibres

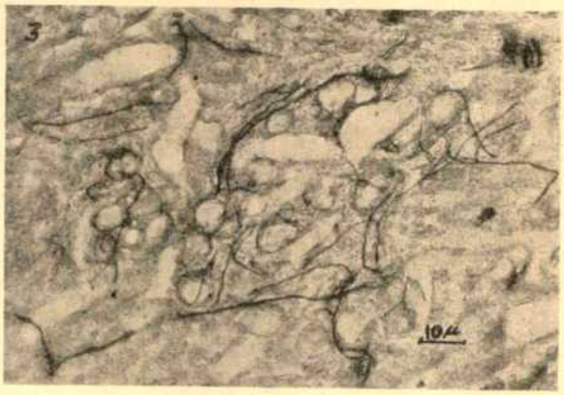

Fig. 3. Part of the carotid body showing the large globular glomus cells, sinusoids, and large nerve fibres running between the glomeruli

One of the difficulties encountered in this study was that the smallest nerve fibres are very near in size to the limits of resolution of the light microscope. However, further support for these observations that : (1) only the Type I glomus cells are directly innervated by the glossopharyngeal fibres without the intervention of interstitial cells, and (2) that no evidence could be found for an innervation of De Kock's Type II cells, comes from the recently made electron microscope studies on the carotid body of the cat by Garner and Duncan[18].

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[1] Berckelmann, M. L. R., "De Nervorum in Arterias Imperio" (1744).

[2] Luschka, H., *Arch. Anat. Physiol. Lpz.*, 405 (1862).

[3] De Castro, F., *Trab. lab. Invest. biol. Univ. Madrid*, **26**, 365 (1926).

[4] Sato, S., *Igaku kenkyuu, Fukuoka*, **6**, 707 (1932). German abstract in *Jap. J. Med. Sci., Anat.*, **5**/ii: abstracts 82 (1934).

[5] Theile, F. W., *Arch. Anat. Physiol., Lpz.*, 419 (1852).

[6] Tandler, J., *Denkschr. Akad. Wiss. Wien*, **67**, 677 (1899).

[7] Danielopolu, D., and Manescu, E., *Z. ges. exp. Med.*, **63**, 143 (1928).

[8] Botar, J., *C.R. Ass. Anat.*, 27th Reunion, 177 (1932).

[9] Botar, J., *Arch. Mus. Hist. Nat. Paris*, Ser. 6, **12**, 243 (1935).

[10] Ask-Upmark, E., *Acta psychiat., Kbh.*, Supp. 6, 1 (1935).

[11] "The Microtomist's Vade Mecum", 528 (1950).

[12] Polley, E. H., *Anat. Rec.*, **125**, 509 (1956).

[13] De Kock, L. L., *Nature*, **167**, 611 (1951).

[14] Meijling, H. A., *Acta Neerl. Morph.*, **1**, 193 (1938).

[15] De Kock, L. L., *Acta Anat.*, **21**, 101 (1954).

[16] De Castro, F., *Acta Physiol. Scand.*, **22**, 14 (1951).

[17] Botar, J., *Arch. Mus. Hist. Nat. Paris*, Ser. 6, **12**, 243 (1935).

[18] Garner, C. M., and Duncan, D., *Anat. Rec.*, **130**, 691 (1958).

HÆMATOLOGY

Some Red-Cell Antigens of *Bison bonasus* as compared to Red-Cell Antigens of Domestic Cattle

RED cells of a bison (*Bison bonasus*) were submitted to serological examination in order to find antigenic similarity and differences between the erythrocytes of bison and those of domestic cattle. The blood groups phenotype of the individual in study was as follows :

$$A - A', E'_1 - W, X_2 - F - J - L$$

(The phenotype was determined using 38 bovine blood-typing reagents :

A ; B, G, I, E_1, E_2, O_1, O_3, Q, T_1, Y, A′, D′, E'_1, E'_3, J′, I′, K′ ; C_1, C_2, X_1, X_2, X_3, W, L′ ; F, V ; J ; L ; M ; S_1, S_2, U_1, U_2, U′ ; Z, ZZ ; Z′.

These reagents were kindly given to us by the Department of Genetics, University of Wisconsin, and by the Institute of Animal Breeding, Royal Agricultural College, Uppsala.)

The red cells of the bison studied were injected into seven rabbits and two heads of cattle (twins). The blood group phenotypes of the immunized animals were as follows :

No. 20/57 – I, Y, E'_3, K′ – C_2, X_3 – F – S_2
No. 21/57 – I, Y, E'_3, K′ – X_3 – F

(For determination of these phenotypes 19 standard reagents—A ; B, G, I, Y, K′ ; C_2X_2, L′ ; F, V ; J ; L ; S_1, S_2, U_1, U_2 ; Z—were used.)

After one series of immunizations (10 injections into each rabbit and three injections into each of the cattle) the animals were bled. The rabbit-immune sera reacted in high titres with both bison and cattle red cells (Table 1). The two cattle-immune sera exhibited the lytic titres of 1 : 64 and 1 : 16 against the bison red cells. These sera contained also antibodies for some samples of cattle red cells.

Table 1. Results of the Agglutination and Lysis Tests, using Unabsorbed and Absorbed Rabbit Anti-bison Red-Cell Sera with Bison and Cattle Red Cells

Serum		Titres of red cells -			
		Bison		Cattle	
		Agglutination	Lysis	Agglutination	Lysis
No. 7272	Unabsorbed	2,048	4,096	0	1,024
	Absorbed	32	64	0	0
No. 7144	Unabsorbed	2,048	2,048	0	512
	Absorbed	16	16	0	0
Pooled serum of all 7 rabbits	Unabsorbed	8,192	8,192	0	1,024
	Absorbed	64	128	0	0

Serum No. 20/57 reacted with 20 of the 110 tested cattle red-cell samples ; serum No. 21/57 reacted with 45 of the 109 tested samples. The specificity of these antibodies could not be identified with any of the 38 standard reagents ; more detailed studies on this problem are in progress.

In order to test whether the bison red cells bear antigenic substances particular only to this species, the rabbit anti-bison red cell immune sera were submitted to absorption with cattle red cells. It was assumed that this absorption would remove antibodies for those bison antigens which are identical or very similar to some of the cattle red-cell antigens.

The 10 samples of the cattle red cells employed in the absorption contained all known antigens except the antigens : H, I, P, E_2 and Z'. In the given case, only the absence of H and P antigens was of consequence. The remaining antigens, that is, I, E_2 and Z', were not present in the bison blood sample, and therefore there was no possibility that any antibodies could be formed against these antigens.

The rabbit anti-bison red-cell sera absorbed with cattle red cells gave a negative reaction with each of the 48 cattle red-cell samples, whereas they gave a clearly positive reaction with the red cells of the bison, and what is more, even in rather high dilutions. These results are presented in Table 1.

Despite the fact that these tests require further development and repetition on a wider range of specimens, our results allow us to conclude that :

(1) The erythrocytes of bison blood contain the antigens : A of system A, $A'E'_1$ of system B, WX_2 of system C, F of system FV, J^{sc} of system J, L of system L. They are all either identical or very similar to the antigens present in the domestic cattle.

(2) The red cells of the tested bison contain other, still unidentified antigenic factor(s) which may perhaps be found also in some of the cattle red-cell samples; these factors are antigenic to those individuals of cattle which do not contain them.

(3) The red cells of the tested bison contain one or more antigenic properties specific for them, which were not found in the cattle red-cell samples studied

Thanks are due to Profs. M. R. Irwin and W. H. Stone of the University of Wisconsin for their friendly encouragement of this research and personal suggestions, to Dr. S. Dubiski of the Institute of Hæmatology, Warsaw, for valuable advice and comments, and to S. Wezyk for handling part of the field work.

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Haptoglobin Types in South-East Asia

The discovery that the haptoglobin present in human serum can be resolved into a number of different types by electrophoresis in starch gel[1] and the subsequent demonstration that these types are controlled genetically[2] has opened a new field of investigation in human population genetics. Already the distribution of the haptoglobin phenotypes has been reported for a number of different populations in the world[3], and several other investigations privately reported to us are in progress.

The present study covers the distribution of the haptoglobin types among the three major ethnic groups in Malaya. Blood samples were obtained by venepuncture from healthy adults. In Perlis the samples were collected into Bayer 'venules' and sent to Perth as clotted blood : in Kuala Lumpur the samples were collected into syringes, and the serum was separated from the clot before being sent to Perth. In both cases the samples were flown in ice-cold containers and were generally in Perth within 48 hr. from the time of collection.

During the early part of the investigation electrophoresis was carried out in horizontal trays according to the original method of Smithies[4], using the hydrolysed starch supplied by the Connaught Medical Research Laboratories and borate buffer. Hæmoglobin was added to the samples prior to electrophoresis to give a concentration of approx-

Table 1 Number of Persons by Haptoglobin Type in various South-East Asian Populations

Place	Ethnic Group	*Hp* type					Total
		0–0	1–1	2–1	2–1 (mod)	2–2	
Perlis	Malays	0	5	53	0	99	157
	Indians	1	2	17	2	110	132
Kuala Lumpur	Malays	2	7	31	1	38	79
	Chinese	1	9	36	0	55	101
	Indians	3	2	13	0	69	87
Perth (students)	Chinese	1	9	21	—	35	66

Table 2

Ethnic group	No. tested	Distribution (per cent)					Gene frequency		$\chi^2(1)$	P
		0–0	1–1	2–1	2–1 (mod.)	2–2	Hp^1	Hp^2		
Malays	236	0·8	5·1	35·6	0·4	58·1	0·24	0·76	0·2	0·7–0·5
Chinese	167	1·2	10·8	34·1	—	53·9	0·28	0·72	3·58	0·1–0·05
Indians	219	1·8	1·8	13·7	0·9	81·7	0·09	0·91	0·70	0·5–0·3

imately 100 mgm. hæmoglobin/100 ml. of serum. The haptoglobin bands were rendered visible by means of a benzidine reagent. In the latter part of the investigation electrophoresis was carried out in vertical trays[5], the other conditions remaining the same.

Table 1 records the haptoglobin types for the Perlis and Kuala Lumpur samples together with a smaller number of samples collected from Asian students in Perth. There is no significant heterogeneity for any of the ethnic groups with respect to the place from which the samples came. For the calculation of gene frequencies the samples have been combined and the 0–0 and *Hp* 2–1 (mod) phenotypes excluded from the calculation.

Several points of interest arise when the present results, as shown in Table 2, are compared with those reported for other populations. In this material the modified *Hp* 2–1 phenotype[6] is rare, and no cases were noted among the Chinese, only two cases in Indians, and a single case in Malays. Similarly, individuals completely lacking haptoglobin (*Hp* 0–0) are infrequent also, though more common than in a control population of Australian whites[7]. It is extremely difficult to distinguish the *Hp* 0–0 phenotype with certainty. In many cases the amount of haptoglobin present in *Hp* 2–2 individuals is so slight that it can readily escape detection. All cases where haptoglobin was missing were examined at least three times on successive days. But some of the *Hp* 0–0 individuals may still represent *Hp* 2–2 individuals with haptoglobin concentrations below the level necessary for detection. Other cases may be individuals temporarily lacking haptoglobin due to hæmolytic episodes in their past history[8].

It has been suggested that the *Hp* 0–0 phenotype may be due to a modifying gene at a separate locus[9], or alternatively represents the homozygous condition for the Hp^{2M} gene (Giblett, E. R., personal communications). We do not have family material which would help in the interpretation of this phenomenon, but in the latter case since the modified *Hp* 2–1 type is infrequent in our series it would suggest that the homozygous condition Hp^{2M}/Hp^{2M} should be rare in these three ethnic groups.

The chief point which emerges from the present study is the high frequency of the Hp^2 gene in this area, reaching its highest reported value in Indians. This result is in agreement with the value reported by Sutton for a group of Asiatic Indian students in North America[10]. Further studies on populations in the Near and Middle East and in central Asia will be necessary before a clear picture of the *Hp* gene distribution in Asia as a whole is obtained. We are extending our own investigations to cover the aboriginal populations in this area, and a survey of the variations in the *Tf* and *Gm* serum protein groups for all these populations in South-East Asia is at present in progress.

Our thanks are due to the Australian National Health and Medical Research Council and the Research Grants Committee of the University of Western Australia, for financial assistance, and to Dr. Eloise Giblett for a sample of a *Hp* 2–1 (mod) serum.

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[1] Smithies, O., *Nature*, 175, 307 (1955).

[2] Smithies, O., and Walker, N. F., *Nature*, 176, 1265 (1955).

[3] Allison, A. C., Blumburg, B. S., and ap Rees, *Nature*, 181, 824 (1958). Sutton, H. E., Neel, J. V., Binson, G., and Zuelzer, W. W., *ibid.*, 178, 1287 (1956). Giblett, E. R., *ibid.*, 183, 192 (1959). Rodriguez, M. L. G., and Arends, T., *ibid.*, 183, 1465 (1959). Harris, H., Robson, E. B., and Siniscalco, M., Ciba Symposium on Medical Biology and Etruscan Origins, 220 (1959).

[4] Smithies, O., *Biochem. J.*, 61, 629 (1955).

[5] Smithies, O., *Biochem. J.*, 71, 585 (1959).

[6] Connell, G. E., and Smithies, O., *Biochem. J.*, 72, 115 (1959).

[7] Kirk, R. L., Lai, L. Y. C., and Hogben, D. L., *Med. J. Austral.* (in the press).

[8] Laurell, C. B., and Nyman, M., *Blood*, 12, 493 (1957)

[9] Harris, H., Robson, E. B., and Siniscalco, M., *Nature*, 182, 1324 (1958).

[10] Sutton, H. E., Neel, J. V., Livingstone, F. B., Binson, G., Kunstadter, P., and Trombley, L. E., *Ann. Hum. Genet.*, 23, 175 (1959).

Retention of Potassium by Human Erythrocyte Ghosts

When ghosts prepared from human erythrocytes by lysis in water are washed in a saline medium, sodium and potassium ions are nearly uniformly distributed across the ghost membranes[1]. In attempting to prepare ghosts with higher potassium and lower sodium concentrations than those of a saline medium we have found that incubation at 37° C. enables them partially to retain potassium and keep out sodium during subsequent washings and incubation.

A hæmolysate of completely lysed human erythrocytes was made approximately isotonic with plasma by the addition of 3 *M* salt solution (5 parts potassium chloride and 1 part sodium chloride) to give final concentrations of 150 m*M* potassium chloride and 30 m*M* sodium chloride. The ghosts were washed three times at room temperature in a saline medium (0·17 *M* sodium chloride, 0·005 *M* potassium chloride, 0·01 *M* sodium phosphate, *p*H 7·4), this process taking about 30 min. The potassium concentration in the ghosts (hæmoglobin content about 10 per cent of original erythrocytes) was approximately three times that in the medium, and incubation of the hæmolysate at various temperatures and for different periods of time before the addition of 3 *M* salt solution made no difference. However, when the suspension was incubated for about 30 min. at 37° C in 150 m*M* potassium chloride after the addition of

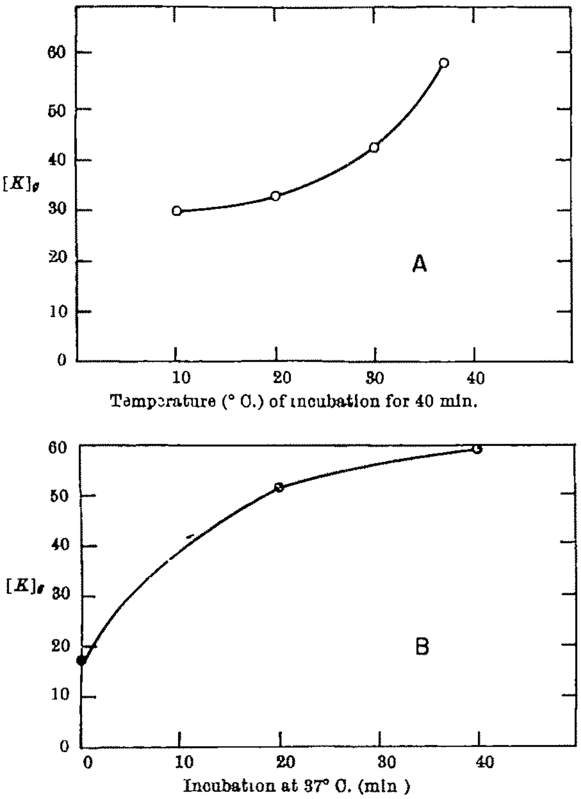

Fig. 1. The effect of incubation of a hæmolysate after the addition of 3 M salt solution to give 150 mM potassium chloride and 30 mM sodium chloride, at different temperatures for 40 min. (graph A), and at 37° C for varying periods of time (graph B), on the potassium concentration of red cell ghosts afterwards washed three times in physiological saline solution. $[K]_g$ has the unit. m.moles potassium/litre of ghosts

the 3 *M* salt solution, the washed ghosts then contained about twelve times more potassium than the medium. Both the temperature and the period of incubation affected the amount of potassium found in the washed ghosts (Fig. 1). The more potassium present the higher the temperature of incubation (range 10–37° C.), and ghosts incubated for 40 min. at 10° C. contained twice as much potassium as those unincubated at any temperature.

Ghosts spun down from a hæmolysate and then incubated in 0·17 *M* potassium chloride acquired a similar ability to retain potassium, showing that the non-particulate part of the hæmolysate was not required. The final potassium concentration of washed ghosts, however prepared, was never more than 50 per cent of the concentration in the suspension before washing. This may be because half the ghosts were equilibrated with the washing medium or because all the ghosts retained only half the maximum possible potassium, although these explanations are not exclusive. During the incubation of washed ghosts in the saline medium for 4 hr. at 37° C., about 5 m.moles potassium/l. ghosts/hr. were lost, which is about twice the rate of loss of potassium from fresh starved erythrocytes.

The results show that ghosts containing about 10 per cent of the hæmoglobin of erythrocytes may partially retain potassium during three washes and subsequent incubation in a saline medium, suggesting that after osmotic hæmolysis ghosts may regain some degree of the low permeability to potassium which is characteristic of erythrocytes.

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[1] Ponder, E., "Hæmolysis and Related Phenomena" (Churchill, London, 1948)

Porphyrin Biosynthesis in the Leucocyte

THE presence, in various animal tissues not primarily hæmatopoietic, of some or all of the enzyme systems taking part in porphyrin and hæm biosynthesis has been reported by many authors. However, until recently, any investigation of porphyrin biosynthesis in the leucocyte has been hindered by the contamination of tissue preparations with erythrocytes having a much greater synthetic capacity. During a systematic study of several enzyme systems in the normal and pathological leucocyte now being carried out in this clinic, a method of preparing undamaged leucocytes, completely free of erythrocytes and thrombocytes, has been developed[1]. It therefore seemed interesting to study the behaviour of these enzyme systems in this readily available and easily quantitated tissue.

After incubation of a homogenate of normal human leucocytes with δ-amino-lævulinic acid under standard conditions (4 hr. at 37° C., *p*H 7·4, 0·8 × 10^7 leucocytes/400 μl.), a substance giving a red colour insoluble in chloroform on reaction with *p*-dimethylaminobenzaldehyde is found in the deproteinized supernatant. The absorption spectra of this colour and that formed by a solution of porphobilinogen in the incubation medium are exactly superposable between 450 and 600 mμ, with peaks at 522 and 555 mμ. No fluorescence is seen under Woods light and only a very slight peak between 400 and 410 mμ. After heating the deproteinized supernatant at 100° C., and *p*H 1–2, for ½ hr., a distinct red fluorescence is seen, together with the appearance of a peak at 407 mμ, indicating strongly that the Ehrlich reacting substance present was porphobilinogen, transformable to porphyrin. A quantitative analysis of these results will be published elsewhere.

It is interesting to note that leucocytes prepared from the blood of normal guinea pigs showed synthesis of porphyrin after 4 hr. incubation. Due to the small amounts no analysis or purification of any porphyrins formed has been attempted. The concentrations were roughly compared by comparison of the corrected absorption at the Soret peak, that is, of the deproteinized incubate supernatants.

Chromatography of the supernatant of an incubate of authentic porphobilinogen with normal leucocytes under standard conditions showed two spots, one corresponding to the porphobilinogen marker together with another, remaining near the origin and becoming blue on standing. Attempts at chromatography of the supernatant from an incubation with δ-amino-

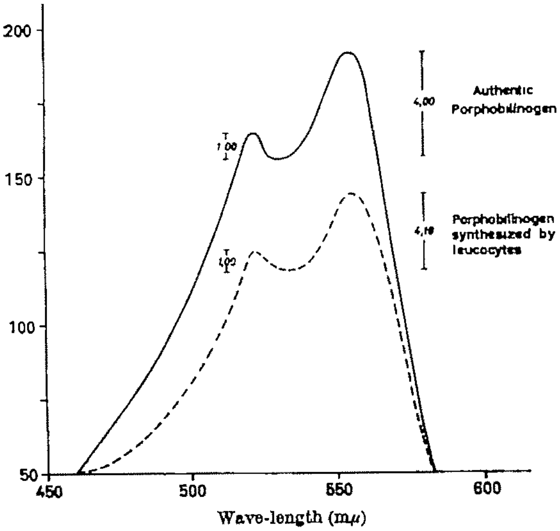

Fig. 1

lævulinic acid were not reliable due to the contamination of the minute quantities involved. However, while no typical porphobilinogen spot was seen, one remaining near the origin and turning blue on standing was present. Demonstration either that this spot is similar to the atypical spot on the porphobilinogen chromatograms or is only an artefact must wait until very much larger quantities are available.

If the incubation is continued, porphyrin fluorescence is seen in the supernatant after 24 hr., indicating that its absence earlier in the incubation could be a problem of the concentration of the different factors involved. However, incubation of a more concentrated homogenate, approx. 40×10^6 leucocytes/incubation, led to synthesis of proportionately greater quantities of porphobilinogen, always without the appearance of porphyrin. Incubation of authentic porphobilinogen in physiological quantity (6 mμM) with leucocytes under standard conditions leads to porphyrin formation apparently in excess of that in the control without leucocytes; but as this remains constant when either the concentration of porphobilinogen or of leucocytes is increased, the apparent difference is probably due to the method of estimation. It seems most likely that the presence of porphyrin after 24 hr. incubation is due to the spontaneous condensation of the porphobilinogen synthesized, and that the mature leucocyte, while able to form porphobilinogen, cannot carry the synthesis any further. There are two possible explanations of this: that porphobilinogen-deaminase is in fact absent, or that it is present but inhibited in some fashion.

That it is present and active in the immature cell is indicated by the results of incubation of leucocytes from certain cases of leukæmia. When the cell preparation contained a large proportion of young cells, either monoblasts, lymphoblasts or myeloblasts, there was net synthesis of porphyrin after incubation with δ-amino-lævulinic acid under standard conditions. It is possible that, on maturation of the leucocyte, a system which slightly transforms porphobilinogen so that it cannot be utilized by porphobilinogen-deaminase may come into play.

It is also interesting to note that in two cases, one of acute myeloid and one of acute lymphatic leukæmia where the cells contained little or no protoplasm, there was no synthesis of either porphobilinogen or porphyrin from δ-amino-lævulinic acid, demonstrating that these enzymes are probably exclusively present in the cytoplasm.

We wish to thank Prof. C. Rimington for his advice on our chromatograms

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[1] Borel *et al.*, *Acta Med. Internisten*, 063/31 (1959).

Hæmolytic Disease of the Newborn probably due to Anti-f

A case is briefly presented, the chief feature of which is the production of anti-f held to be responsible for severe hæmolytic disease of the newborn. Prior to her marriage the patient, Mrs. M. H., group O, Rh positive, most likely genotype R^1R^1 (DCe/DCe), was transfused with Rh positive blood of unknown genotype. The first pregnancy ended in a full-term stillbirth but no autopsy was performed. The second pregnancy also terminated in a full-term stillborn due to hydrops. During this second delivery the patient received 500 c.c. of group O, Rh negative blood followed by a severe hæmolytic reaction and renal shutdown. The patient recovered with the aid of an artificial kidney and received a total of 13 pints of blood—all compatible by an indirect Coombs cross-match.

The third infant delivered in 1958 by Cæsarean section at 38 weeks had severe hæmolytic disease and recovered after replacement transfusion with group O, Rh positive compatible blood.

The first blood specimen of Mrs. M. H. was submitted during August 1959 in the seventh month of her fourth pregnancy. Screening tests indicated that the serum contained an antibody which reacted with albumin-suspended cells. On testing the serum with a panel of cells it was observed that bloods R^1R^1(DCe/DCe), R^1R^2(DCe/DcE), $r'r'$(dCe/dCe) and r^yr^y(dCE/dCE) failed to react, while R^2R^2(DcE/DcE) gave either negative or very weak reactions. Cells of genotypes R^1r and rr and others having antigens c and e carried by the same chromosome in single or double dose reacted strongly. The failure of the serum to react with the 4 $r'r'$ and r^yr^y bloods excludes the presence of anti-d. These findings suggested the presence of a very weak anti-c but a predominating antibody, anti-f. In order to exclude a prozone indicative of a potent anti-c, the serum was titrated with R^2R^2 cells and this result confirmed the presence of a very weak anti-c. The titre of anti-f as determined by tests with rr bloods was 1 : 32. These findings were fully confirmed in tests of a second specimen of the patient's serum submitted later in the seventh month

Mrs. M. H. was delivered by Cæsarean section one month prior to the expected date of delivery and the baby was reported as normal at birth, but soon developed jaundice. The direct Coombs test of the cord blood was moderately positive. The infant

Table 1. Tests of the Red Cells of the Parents and Affected Infant

		D	C	E	c	e	f	K	k	Fy^a	S	s	M	N	P
Father	O	+	+	0	+	+	+	0	+	0	0	+	0	+	+
Mother	A	+	+	0	0	+	0	+	+	+	+	0	+	0	+
Cord	A	+	+	0	+	+	+	Not tested							

Table 2

Probable genotype	No. of tests	Reactions with anti-f	Maternal serum		Cord serum		Eluate cord red cells	
			1	2	1	2	1	2
R^1R^1	3	0	0	0	0	0	0	0
R^1R^2	1	0	0	0	0	0	0	0
R^2R^2	4	0	0	0	0	0	0	0
R^2R^2	1	0	+	0	0	0	0	0
R^1r	2	+	++++	++++	++	++++	0	++++
R^2r	1	+	++++	++++	+++	++++	0	++++
R^0r	1	+	++++	++++	+++	++++	0	++++
$r'r$	2	+	++++	++++	+	++++	0	++++
$r''r$	2	+	++++	++++	++++	++++	0	++++
rr	4	+	++++	++++	++++	++++	0	++++
$r'r'$	4	0	0	0	0	0	nt	nt
$r'r''$	1	0	0	0	nt	nt	nt	nt
r^yr^y	1	0	0	0	0	0	nt	nt
R^{oz} $(D--/D--)$	4	nt	0	0	nt	nt	nt	nt

nt, not tested.

Column 1 indicates the reactions with albumin-suspended red cells after centrifugation; column 2 indicates the reactions with indirect antiglobulin test on the albumin-suspended red cells.

The anti-*f* and $r'r''$ bloods were supplied by courtesy of Dr. R. E. Rosenfield, Blood Bank, Mount Sinai Hospital, New York.

recovered following a replacement transfusion carried out with Rh positive blood, compatible with the mother's serum.

An eluate prepared from the cord red cells by the heat technique, revealed the presence of anti-f when tested with albumin-suspended cells followed by the indirect antiglobulin reaction. The results obtained in simultaneous tests of the maternal serum, cord serum and the eluate of the infant's coated red cells with a selected panel of test cells are given in Table 2.

Since the supply of maternal serum obtained at delivery was exhausted, the specimen taken at 7 months was compared with cord serum in parallel titrations with ficinated red cells. The results are given in Table 3.

Table 3

Test cell	Maternal serum	Cord serum	Test for
R^2R^2	1:4	1:4	anti-c
rr	1:16	1:32	anti-f

The titre for anti-c was less than the titre for anti-f, and this observation together with the findings of the cord cell eluates supports the view that anti-f induced the hæmolytic disease. This case is unique because the patient was expected to produce the more common anti-c and perhaps anti-f. Surprisingly, anti-c was very weak and a more potent and readily detectable anti-f was produced.

In the first example of anti-f, produced by a patient immunized by transfusions, there were additional antibodies (anti-N, anti-K and anti-S), but anti-f was the only Rh antibody produced[1,2]. Anti-f is frequently found as a secondary antibody in most anti-c and anti-e sera[3], but its demonstration is not a simple matter and requires absorption with selected cells and testing with enzyme-treated cells.

There is no apparent explanation as to why this mother who should have produced the more usual combination of a strong anti-c and perhaps some anti-f responded preferentially to the 'weaker' antigen. Studies show nothing unusual in the genotype of her husband's blood. His c antigen scored the same as other R^1r bloods. The antigenic stimulus in this case, however, was provided prior to her first pregnancy by transfusions of bloods which were probably f positive and during the delivery of the second stillbirth by Rh negative, and, therefore, f positive blood. There is in the literature at least one example of a patient immunized by pregnancy who produced the rare instead of the more common antibody[4]. In this case an Rh negative patient produced a more potent anti-C and a much weaker anti-D. Her husband was of the unusual genotype R^1r' (DCe/dCe) with suppression of D by gene interaction with the partner chromosome r' (dCe). We and others have also observed several examples of Rh negative women who produced anti-K or other antibodies and not anti-D even though their husbands were Rh positive.

We wish to thank Dr. Robert J. Collins, Snyder, New York, for submitting these specimens and supplying us with the case history.

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[1] Rosenfield, R. E., *et al.*, *Brit. Med. J.*, **1**, 975 (1953).
[2] Sanger, R., *et al.*, *Proc. U.S. Nat. Acad. Sci.*, **39**, 824 (1953).
[3] Race and Sanger, "Blood Groups in Man", 3rd edit.
[4] Levine, P., *et al.*, "Andresen Dedication Book", 144 (Munksgaard, Copenhagen 1957).

PATHOLOGY

Auto-Antibody in Rats with *Plasmodium berghei*

IN human malaria it has long been recognized that total loss of blood is significantly more extensive than loss which can be attributed to the direct rupture of parasites emerging from infected erythrocytes[1]. In a comparative survey of loss of blood and replacement in plasmodial infections of other mammals and birds, excessive erythrocyte destruction of the same type has been observed in all except one of the host–parasite combinations studied to date, namely in rats with *Plasmodium berghei*[2] and *Plasmodium vinckei*[3], in rhesus monkeys with *Plasmodium cynomolgi*, *Plasmodium gonderi* and *Plasmodium knowlesi*[4], and in chickens with *Plasmodium lophurae* (Zuckerman, A., unpublished results).

In this survey excessive loss was particularly striking in infections with mild or negligible parasitæmia. In such cases the main anæmic bout generally follows peak parasitæmia by at least a few days, and usually occurs when parasitæmia has become microscopically negligible or even latent. Since direct rupture due to emerging parasites cannot but be negligible when parasitæmia is so low; and while during the same period up to a third or even more of the circulating red cells may rapidly be destroyed, the results clearly show that some agency other than direct parasitic rupture is engaged in destroying red cells.

A working hypothesis to explain this excessive loss was suggested in the earlier papers of the survey, namely, that red cells may be sufficiently altered by the presence of parasites to become antigenic, and that they may then give rise to the production of auto-antibody.

The host–parasite combination of *P. berghei* in the adult rat is a convenient one for demonstrating excessive loss of blood, since parasitæmia is low and loss of blood high. Excessive loss thus stands out clearly, and is not masked by loss due to the parasitic rupture of host cells.

Auto-antibody was sought in such rats with the aid of a modified Coombs's test using anti-rat-serum rabbit serum as antiserum. The antiserum, as well as normal control rabbit serum, was exposed to normal washed rat erythrocytes before use, to remove an anti-rat-cell agglutinin regularly present in rabbit serum.

Forty-nine Coombs's tests done in 14 infected rats were positive during the period following parasitic crisis, while Coombs's negatives were regularly obtained from the same rats before crisis. To date, no infected rat which survived until latency has failed to pass through an evanescent period during which its red cells were antibody-coated. In no case did agglutination of Coombs's positive red cells occur in the presence of normal adsorbed rabbit serum. Forty-five tests done with the blood of eight uninfected control rats exposed to normal or anti-rat-serum rabbit serum were all Coombs's negative.

Antibody coating of erythrocytes, negative before crisis and positive after crisis, wanes and finally disappears as blood counts return to normal. The period of antibody coating thus closely corresponds to that of excessive loss of blood. Since latency commences at about the same time, the overwhelming majority of the coated cells must have been uninfected rather than parasitized.

Removal of the antibody-coated cells from the circulation is thought to be the cause of the excessive anæmia.

Bilirubin was sought in the serum of four rats at the height of the anæmic bout, with negative results. Antibody coating at this time is therefore not likely to have led to intravascular hæmolysis. It is more likely that coated cells, being opsonized, are filtered out of the circulation by elements of the lymphoid–macrophage system. It is germane that unparasitized red cells, together with infected cells, are avidly taken up at and after crisis by macrophages of monkeys infected with *Plasmodium brasilianum*[5], *P. cynomolgi* and *P. knowlesi*[6]; and that chicken macrophages *in vitro*, in the presence of blood infected with *P. gallinaceum* or *P. lophurae* suspended in homologous hyper-immune serum, ingest both normal and infected cells[7].

The above experiments are compatible with the view that auto-antibody against their own uninfected red cells is formed in mature rats infected with *P. berghei*. The fact that excessive loss of blood similar to that in *P. berghei* rats has also been observed in a broad spectrum of plasmodial infections in hosts of widely varying phylogeny suggests that a similar process may prove to be a general feature of malarial pathology.

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[1] Maegraith, B., "Pathological Processes in Malaria and Blackwater Fever", 75 (Blackwell Scientific Pub., Oxford, 1948).

[2] Zuckerman, A., *J. Inf. Dis.*, 100, 172 (1957).

[3] Zuckerman, A., *J. Inf. Dis.*, 103, 205 (1958).

[4] Zuckerman, A., *J. Inf. Dis.* (in the press).

[5] Taliaferro, W. H., and Cannon, P. R., *J. Inf. Dis.*, 59, 72 (1936).

[6] Taliaferro, W. H., and Mulligan, H., *Ind. Med. Res. Memoir*, 29, 1 (1937).

[7] Zuckerman, A., *J. Inf. Dis.*, 77, 28 (1945).

Anti-tumour Activities of Copper Chelates

CHELATING agents have been shown to interfere with the utilization of metals by micro-organisms, robbing cells of their necessary metals[1]. Retardation of growth of a tumour (sarcoma 37) by the administration of two chelating agents, dipyridyl and O-phenanthroline, was observed by Leiter *et al.*[2]. Now, α-ketoglutaric acid oxime has been found to form its copper, iron, or nickel chelate at the physiological *p*H, and its antitumour activity *in vitro* was observed by the stained cell count method[3].

The immediate injurious action of the oxime (100 mgm./ml.) on the selective permeability of Ehrlich carcinoma cells was shown to be increased by the formation of chelate compounds. The same action of the chelates, copper and iron-α-ketoglutaric acid oxime, was brought about by a concentration of 25 mgm./ml. calculated as the oxime. To show the clear antitumour activity, the hydrophilic groups of the chelating agent should be masked by the metal ion to form the chelate compound which will be more permeable through cell membranes including the lipoid layer[4].

Dimethylglyoxime did not exhibit antitumour activities at a dose of 50 mgm./kgm./day *in vivo*. But the oxime (3–6 mgm./kgm./day), a chelating agent, exhibited potential antitumour activities on Ehrlich carcinoma and Crocker sarcoma 180 in the presence of copper, and not in the co-existence of other metal ions, such as nickel, cobalt, zinc, iron, manganese, magnesium and mercury. If antitumour activity of the chelate compound is only due to that permeable nature, the dimethylglyoxime should also exhibit the activity in the existence of other metals. But the activity was only exhibited by the copper chelate compound.

Not all the copper chelates always exhibit antitumour activity. The ligand moiety may also take a significant part in the activity. If the activity be exhibited by the non-ionic molecule of the copper chelates, copper-biacetyl-monoxime should also exhibit antitumour activity.

Both copper-dimethylglyoxime and copper-biacetylmonoxime inhibited the dehydrogenase activity of Ehrlich carcinoma cells at the same concentration of 25 γ/ml. But the former destroyed the selective permeability of tumour cells at 25 γ/ml.; nevertheless in the case of the latter chelate more than 180 γ/ml. were required to cause such action. As has already been described, the former exhibited clear antitumour activity *in vivo*, while the latter did not. Both of them might permeate the cells to inhibit dehydrogenase activity, and only the former clearly inhibited tumour growth.

De Long *et al.*[5] observed a decreased calcium content which is peculiar to tumour tissues. Ambrose *et al.*[6] observed a marked difference of the electrophoretic motility between tumour and normal cells. As suggested by Brachet[4], if the increased motility of the cancer cells is to be attributed to different properties of the cell membrane, the antitumour substances, having specific inhibitory actions by breaking exclusively the selective permeability of cancer cells, may be formed. Some copper chelates having an adequate ligand moiety may exhibit the specific inhibitory action on tumour cells.

A full account of this work including the antitumour activities of the copper-dimethylglyoxime is to be published in the journal *Gann.*

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[1] Forsfall, J. G., "Principles of Fungicidal Action" (Chronica Botanica Co., Waltham, Mass., 1956).

[2] Leiter, J., *et al.*, *J. Nat. Cancer Inst.*, 14, 365 (1953).

[3] Kikuchi, K., and Matsuzawa, M., *J. Antibiotics* (Japan), Ser. B, 8, 170 (1955). Kamata, H., *et al.*, *ibid.*, Ser. B, 11, 1 (1958).

[4] Brachet, J., "Biological Cytology" (Academic Press, Inc., 1957).

[5] De Long, R. P., Coman, D. R., and Zeldman, I., *Cancer*, 3, 718 (1950).

[6] Ambrose, E. J., James, A. M., and Lowich, J. H. B., *Nature* 177, 576 (1956).

'Phenergan' and Versene in Dietary Liver Necrosis

RATS fed on Himsworth's[1] necrogenic diet have livers that are histologically undamaged, except for mild fatty infiltration, until an episode of acute massive necrosis occurs. Schwarz *et al.*[2] reported that liver slices from animals fed a necrogenic diet showed a metabolic abnormality at times when the livers were still normal histologically. The abnormality consists of an inability of these slices to maintain oxygen uptake for more than about 30 min. when incubated in Ringer solution at 38° with oxygen.

In the present investigation acute massive liver necrosis overtook the weanling rats after about fourteen days of feeding the yeast diet (mean 17·5, *S.D.* 4·0). Slices were taken from rats without liver necrosis at times after the seventh day of feeding the diet. Oxygen uptake was measured by the Warburg technique with Ringer phosphate[3] solution with 1·2 m*M* calcium as the incubation medium. Respiration declined to 50 per cent of initial rates in 40–60 min. This decline was invariably and completely prevented over test periods of 150 min. by the addition of 10^{-4} *M* 'Phenergan' (10-(2-dimethyl amino-*iso*propyl)phenothiazine hydrochloride) or 10^{-3} *M* versene (sodium ethylenediamine tetraacetate) (Fig. 1).

The initial rate of oxygen uptake in Ringer solution was the same as the rate maintained by the protective additions, and was similar to the rate of oxygen uptake by slices from control rats fed the necrogenic diet plus α-tocopherol (which completely prevents necrosis).

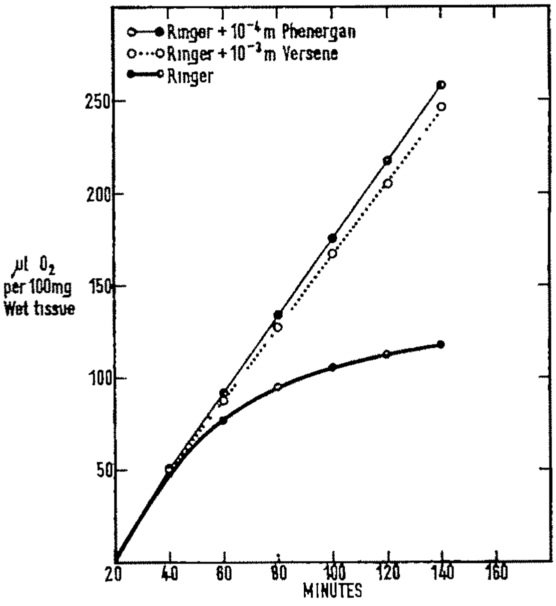

Fig. 1. Oxygen uptake of liver slices. Rat on yeast diet for 9 days

If 'Phenergan' is added to the yeast diet fed to the rats the onset of massive liver necrosis is delayed (Fig. 2).

The same effect is found if the two groups of rats are pair fed, and it is therefore not due to the decrease in food intake associated with the first days of feeding 'Phenergan'.

'Phenergan' has been found to prevent liver damage in thioacetamide poisoning[4]. A possible explanation for these effects in dietary liver necrosis is that both 'Phenergan' and versene act by blocking a chain of reactions where trauma leads to release of toxic breakdown products and further damage results. Such trauma would be provided by cutting the slices, or, *in vivo*, by absorption of toxic substances from the diet. Work is in progress to elucidate the nature of the biochemical lesion.

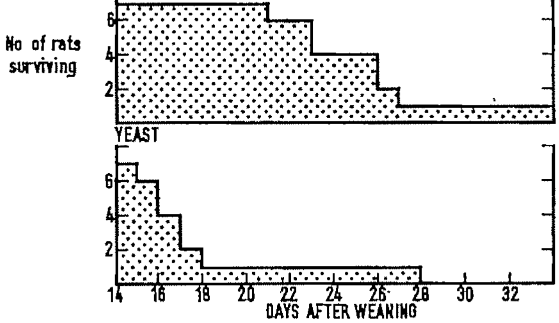

Fig. 2. 21 rats given yeast diet alone for 13 days from weaning. Then divided into three groups and fed yeast diet with or without 'Phenergan'

This work was done during the tenure of a Colonial Medical Research Scholarship.

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[1] Lindan, O., and Himsworth, H. P., *Brit. J. Exp. Path.*, 31, 651 (1950).

[2] Chernick, S S , Moe, J. G., Rodnan, G. P., and Schwarz, K., *J. Biol. Chem.*, 217, 829 (1955).

[3] Cohen, P. P., in "Manometric Techniques and Tissue Metabolism", ed. Umbreit, 119 (Burgess, Minneapolis, 1949).

[4] Gallagher, C. H., Gupta, D. N., Judah, J. D., and Rees, K. R., *J. Path. Bact.*, 72, 193 (1956).

RADIOBIOLOGY

Radioactive Fission Products in Lungs

We have made measurements on the radioactivity of lung tissue, which show that the amount of fission products at present deposited in lungs by inhalation is large enough to be detected by comparatively simple means. This seems of interest for studying retention of dust in the respiratory tract.

Gamma-scintillation spectra of dust samples collected and measured in Vienna since July 1958 have all shown three characteristic photo-peaks: at (1) 145 keV., (2) 490 keV. and (3) 750 keV. This is in agreement with observations at other locations on top soil[1], rain-water[2], etc. Fig. 1*a* gives the spectrum of one of the dust samples collected in Vienna by filtration of air. On the basis of known half-lives, decay schemes and fission yields, the peaks can be assigned to the following fission products: (1) cerium-141 and cerium-144, (2) ruthenium-103, (3) zirconium-95 + niobium-95. These assignments were confirmed in this laboratory by following the decay of the photo-peaks and by carrying out radiochemical separations. Using the peaks, the listed fission products present in atmospheric dust due to nuclear test explosions can be determined quantitatively by gamma-spectrometry.

Since dust particles are inhaled and some fraction of them is retained in the respiratory tract, we have attempted to detect fission products in lungs by gamma-spectrometry. Lungs from animals and humans were ashed and the gamma-spectrum of the ashes was measured using a well-type sodium iodide (thallium) crystal. The spectrum of the ashes of a human lung is shown in Fig. 1*b*. It is apparent that the fission products detected in dust by gamma-spectrometry are also present in lung tissue. From the spectra the contents of zirconium-95 + niobium-95 and of ruthenium-103 can be calculated. (Contents of radiocerium can also be calculated, but the values are thought to be less reliable and are therefore not listed.) The values obtained from our measurements are given in Table 1.

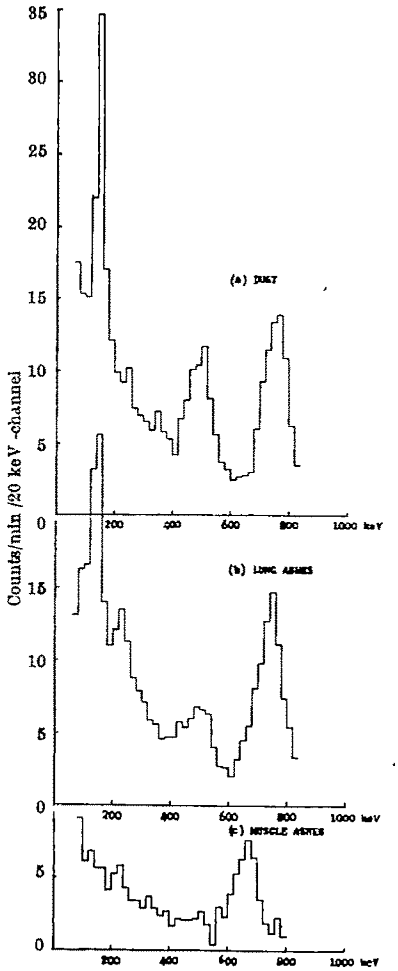

Fig. 1. Typical gamma-spectra (obtained with 20 keV.-channels, counting 16 min. in each channel; background subtracted) (*a*) Dust sample from 88 m.^{3} air (collected April 27, 1959; measured May 6, 1959). (*b*) Lung ashes from human *C* (measured May 26, 1959). (*c*) Muscle ashes from human *C* (measured May 27, 1959)

Table 1. Fission Products in Organs

Organ	Origin of sample	Date of death§	Weight of sample (gm)	Weight of ashes (gm.)	Activity in sample (μμc.)		
					^{95}Zr + ^{95}Nb	^{103}Ru	^{137}Cs
Lung*	cow	24.2.59	4,500	32·1	1,900	280	—¶
Lung*	sheep *A*	6.4.59	310	4·1	150	30	—
Lung*	sheep *B*	6.4.59	340	4·5	240	35	—
Lung*	sheep *C*	13.4.59	320	2 9	175	45	—
Lung*	sheep *D*	13.4.59	290	5 1	140	35	—
Lung*	human *A* (50 years)‡	24.4.59	1,370	8 1	370	55	—
Lung†	human *B* (73 years)‡	12.5.59	400	1·8	155	25	—
Lung†	human *C* (60 years)‡	17.5.29	600	1 9	250	45	—
Muscle	human *B*		710	6·9	—¶	—¶	120
Muscle	human *C*		450	2 6	—	—	100
Liver	human *B*		1,350	12 9	—	—	100
Liver	human *C*		1,605	15 3	—	—	115

* Sample consisted of entire lung.
† Sample consisted of approximately half a lung.
‡ Age at death.
§ All radioactivity measurements were carried out within 5–10 days after death.
¶ Activity not detected or too small for quantitative evaluation.

Control measurements have been made on other organs (muscle, liver) from two of the humans from which lungs were obtained. The spectra of the muscle and liver ashes show only the presence of cæsium-137 (photo-peak at 662 keV.); the characteristic peaks found for dust and lung samples are not observed (Fig. 1c). (The peak due to the 1·46 MeV.-gamma radiation of potassium-40 was also observed, but is not shown in the diagram.) The potassium contents of the ashed lung and muscle tissues from one of the humans were determined by chemical analysis. From the potassium found in muscle ashes a value of the specific activity of cæsium-137 referred to potassium was obtained (84 μμc. cæsium-137 per gram potassium). (This value is somewhat higher than the range of values found by whole-body counting during 1956–57 (25–70 μμc. per gm. potassium, ref. 3) or early in 1958 (30–60 μμc. per gm. potassium, ref. 4).) Using this specific activity and the potassium content of the lung ashes it can be estimated that the activity of cæsium-137 in the lung amounts approximately to one-tenth only of the observed zirconium-95 + niobium-95 activity. This explains why cæsium-137 was not clearly detected in the spectra of lung ashes.

The amounts of radioelements found in lungs can be compared with the corresponding figures for air, as obtained from measurements on dust samples. During the past few months we have usually observed concentrations of fission products within the following ranges: 2·5–5·0 μμc. zirconium + niobium-95 per m.^{3} air and 0·8–1·6 μμc. ruthenium-103 per m.^{3} air. Thus, lungs of adult humans contain at the present time, by order of magnitude, as much fission products as a hundred cubic metres of air. It should also be noted that the activities of fission products found for the various lung samples (cow, sheep and human) agree fairly well if referred to one gram of lung tissue: for the activity of zirconium-95 + niobium-95 six values are within the range 0·39–0·54 μμc. per gram tissue and all eight values are within the range 0·27–0·74 μμc. per gram tissue; for the activity of ruthenium-103 five values are within the range 0·060–0·091 μμc. per gram tissue, and all eight values are within the range 0·04–0·14 μμc. per gram tissue.

The possibilities of using measurements of the type reported here for the study of dust retention and excretion will be discussed in a later detailed publication.

This investigation has been carried out as part of work done under a research contract with the International Atomic Energy Agency. Human lungs were put at our disposal by Prof. H. Hayek, Institute of Anatomy, University of Vienna. The dust samples were collected by Prof. J. A. Schedling, Mr. W. A. Müller and Mr. M. Sedlacek, First Physical Institute, University of Vienna.

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[1] Gustafson, P. F., Marinelli, L. D., and Brar, S. S., *Science*, 127, 1240 (1958).

[2] Schmier, H., *Atomkernenergie*, 3, 346 (1958).

[3] Anderson E. C., Schuch, R. L., Fisher, W. R., and Langham, W., *Science*, 125, 1273 (1957). Report of the U.N. Scientific Committee on the Effects of Atomic Radiation (New York, 1958).

[4] Miller, C. E., Second U.N. Int. Conf. Peaceful Uses Atomic Energy, Report *P* 762 (Geneva, 1958).

Incorporation of Tritium-labelled Thymidine and Tritium-labelled Deoxycytidylic Acid into Deoxyribonucleic Acid after Irradiation of the Deoxyribonucleic Acid Primer with γ-Rays

RECENTLY, Kornberg *et al.*[1] have demonstrated that deoxyribonucleic acid formed *in vitro* by enzymatic synthesis has chemical and physical properties similar to the acid used as primer. Since exposure to radiation is known to cause chemical changes in deoxyribonucleic acid[2–4], it was thought that such changes might be reflected in the newly synthesized acid. In an attempt to study this, we have determined the ability of irradiated and non-irradiated deoxyribonucleic acid to incorporate tritium-labelled thymidine or tritium-labelled deoxycytidylic acid in the presence of the deoxyribonucleic acid-synthesizing enzyme system from regenerating rat liver.

Deoxyribonucleic acid isolated from calf thymus by the method of Kay *et al.*[5] was used in all experiments. An aqueous solution of the acid (0·4 per cent) was irradiated with various doses of γ-rays from a cobalt source at a rate of 4,000 roentgens/min. Irradiated as well as non-irradiated samples were then used as primers to study the incorporation of either labelled thymidine or labelled deoxycytidylic acid into deoxyribonucleic acid in the presence of the following compounds: deoxyadenylic acid, deoxyguanylic acid, deoxycytidylic acid, thymidine, adenosine triphosphate and the 'deoxyribonucleic acid-synthesizing enzyme system' prepared from regenerating rat liver. The assay procedure used was that of Bollum and Potter[6] except that the incubation time was 30 instead of 60 min.

The results of two series of experiments using two tritiated precursors of deoxyribonucleic acid and three radiation doses are shown in Table 1. Each value with standard deviation of the mean is based on quadruplicate or quintuplicate assays. It is apparent that the amount of labelled thymidine incorporated into deoxyribonucleic acid diminished as the radiation dose to the primer increased. In the case of labelled deoxycytidylic acid, however, the amount incorporated increased after irradiation of the deoxyribonucleic acid with 20,000 roentgens but decreased with higher doses to values approximating to those observed with non-irradiated deoxyribonucleic acid. Fig. 1 shows that the ratio of tritiated deoxycytidylic acid to thymidine incorporated into deoxyribonucleic acid was linearly related to the radiation dose under these conditions of study.

Base analysis of irradiated and non-irradiated deoxyribonucleic acid using Wyatt's paper chromatography method[7] showed that irradiation caused no significant change in the base ratio even after exposure to 200,000 roentgens. This indicates that other types of physico-chemical changes in the primer such as those involving hydrogen bonds must be responsible for our observations.

It is interesting to speculate that the changes in the capacity of irradiated deoxyribonucleic acid serving as a primer to incorporate tritiated thymidine and deoxycytidylic acid may be analogous to the altered ability of mutated genes to replicate themselves. A simple calculation serves to show that this hypothesis is not beyond the realm of possibility.

The following reasonable values can be used to calculate the mutation-rate of a gene in an irradiated living cell: (*a*) the molecular weight of deoxyribo-

Table 1. Incorporation of Tritium-labelled Thymidine and Deoxycytidylic Acid into Irradiated Deoxyribonucleic Acid

Group	Experiment 1 Radiation dose (roentgens) 0	 20,000	 240,000	Experiment 2 Radiation dose (roentgens) 0	 20,000	 120,000
Radioactivity* of tritium-labelled thymidine incorporated into deoxyribonucleic acid	159 ± 14	109 ± 15	9 ± 5	64 ± 8	48 ± 4	5 ± 1
Radioactivity* of tritium-labelled deoxycytidylic acid incorporated into deoxyribonucleic acid	41 ± 3	113 ± 25	55 ± 8	70 ± 9	113 ± 10	45 ± 6
Ratio† of incorporation of tritium-labelled deoxycytidylic acid to tritium-labelled thymidine into deoxyribonucleic acid	1·00	4 00	23 5	1 00	2 16	8·90

*Radioactivity is expressed by counts per min. per 100 μgm. of deoxyribonucleic acid.

$$\dagger\ \text{Ratio} = \frac{\left(\dfrac{\text{Radioactivity of tritium-labelled deoxycytidylic acid in irradiated deoxyribonucleic acid sample}}{\text{Radioactivity of tritium-labelled thymidine in irradiated deoxyribonucleic acid sample}}\right)}{\left(\dfrac{\text{Radioactivity of tritium-labelled deoxycytidylic acid in non-irradiated deoxyribonucleic acid sample}}{\text{Radioactivity of tritium-labelled thymidine in non-irradiated deoxyribonucleic acid sample}}\right)}$$

nucleic acid is 6×10^6 (ref. 8); (*b*) one molecule of deoxyribonucleic acid contains 2×10^4 nucleotides, each of molecular weight 300; (*c*) the size of the nucleus is 5×10^{-11} cm.^{3} (ref. 9); (*d*) the deoxyribonucleic acid content per cell is 5×10^{-12} gm. (ref. 10). Assuming that one molecule of deoxyribonucleic acid corresponds to a gene, the mutation-rate in the irradiated cell would be $5{\cdot}8 \times 10^{-5}$ G per roentgen per nucleus per gene, where G equals the number of molecules affected per 100 eV. of energy absorbed. In the experiments reported in this communication, the above-mentioned ratio of 3 : 1 for the tritiated deoxycytidylic acid and thymidine incorporated into deoxyribonucleic acid irradiated with 20,000 roentgens can be used to calculate the G values of this system. This turns out to be 44, provided all nucleotides are equally important in the synthesis of deoxyribonucleic acid.

Substituting this value for G in the expression given above, the rate of molecular alteration of the irradiated deoxyribonucleic acid primer (which could be thought of as analogous to the mutation-rate in a gene) is $2{\cdot}7 \times 10^{-3}$ per roentgen per nucleus per deoxyribonucleic acid molecule. This value is much higher than the mutation-rate of 10^{-4} or 10^{-5} per gene observed in Nature. However, in other studies of irradiated deoxyribonucleic acid such as those involving its transforming activity[11], it has been shown that only one-tenth of the molecule is responsible for the activity. If the primer has a comparably small active site, the rate of alteration of irradiated deoxyribonucleic acid per roentgen would be reduced considerably. When irradiated *in vivo*, protection of deoxyribonucleic acid by other nuclear constituents would also help to reduce the difference between the rates noted in the genes of living cells and those noted under the conditions of our experiments.

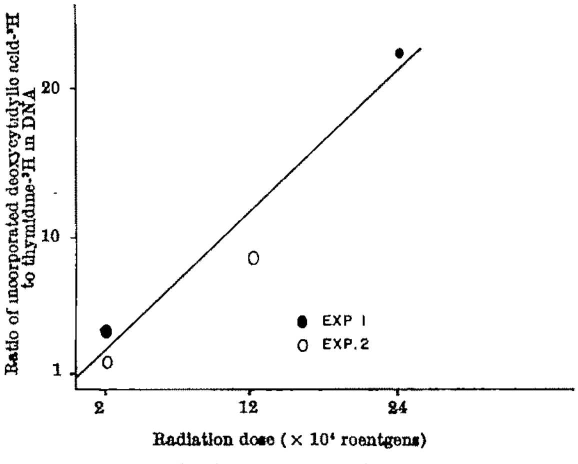

Fig. 1. Changes of ratio of tritium-labelled deoxycytidylic acid to tritium-labelled thymidine incorporated into deoxyribonucleic acid primer irradiated with various doses of radiation. The ratio defined in Table 1 has been used

Further studies on the replicating ability of irradiated deoxyribonucleic acid will be reported elsewhere.

This work was supported, in part, by a grant from the United States Atomic Energy Commission.

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[1] Lehman, I. R., Zimmerman, S. B., Adler, J., Bessman, M. G., Simms, E. S., and Kornberg, A., *Proc. U.S. Nat. Acad. Sci.*, 44, 1191 (1958).

[2] Scholes, G., Stein, G., and Weiss, J., *Nature*, 164, 709 (1949).

[3] Daniels, M., Scholes, G., Weiss, J., and Wheeler, C. M., *J. Chem. Soc.*, 226 (1957).

[4] Cox, R. A., Overend, W. G., Peacocke, A. R., and Wilson, S., *Proc. Roy. Soc.*, B, 149, 511 (1958).

[5] Kay, E. R. M., Simmon, N. S., and Dounce, A. L., *J. Amer. Chem. Soc.*, 74, 1724 (1952).

[6] Bollum, F. S., and Potter, V. R., *J. Biol. Chem.*, 233, 478 (1958).

[7] Wyatt, G. R., "The Nucleic Acids", 1, ed. by Chargaff, E., and Davidson, J. N., 243 (Academic Press, Inc., New York, 1955).

[8] Jordan, D. O., "The Nucleic Acids", 1, ed. by Chargaff, E., and Davidson, J. N., 473 (Academic Press, Inc., New York, 1955).

[9] Dounce, A. L., "The Nucleic Acids", 2, ed. by Chargaff, E., and Davidson, J. N., 123 (Academic Press, Inc., New York, 1955).

[10] Leslie, I., "The Nucleic Acids", 2, ed. by Chargaff, E., and Davidson, J. N., 8 (Academic Press, Inc., New York, 1955).

[11] Defilippes, F. M., and Guild, W. R., *Rad. Res.*, 11, 38 (1959).

ARCHÆOLOGY

Spectrographic Analysis of Romano-British Pottery

Fragments of pottery are the most common signs of human occupation on most archæological sites. Their origin can often be deduced from their shapes and fabrics. There are many cases, however, where this evidence alone is ambiguous, and the work described herewith was undertaken to investigate the possibility of identifying the origin from the concentrations of various minor or trace constituents. Such variations of composition of the ceramic body would tend to reflect the variations in the clay from which it was made[1].

A large number of Romano-British mortaria were analysed spectrographically. This type of pottery is particularly well suited to the purpose as it is very often stamped with a potter's name or mark, and therefore its place of origin can frequently be deduced from the distribution of the stamps. The rim shapes and the fabric also tend to follow regional traditions and provide additional evidence. The mortaria chosen for reference standards were either from kiln sites or were those the origin of which could be deduced from archæological evidence.

Table 1. CONCENTRATION RANGES FOR THE REFERENCE ELEMENTS IN MORTARIA REGARDED AS TYPICAL OF PARTICULAR SITES

Origin	No. of samples	Per cent Na_2O	Per cent MgO	Per cent MnO	Per cent TiO_2	Per cent CaO
Lincoln area	3	$0{\cdot}23^{+0{\cdot}09}_{-0{\cdot}05}$	$0{\cdot}48^{+0{\cdot}02}_{-0{\cdot}01}$	$0{\cdot}01^{+0{\cdot}00}_{-0{\cdot}00}$	$1{\cdot}25^{+0{\cdot}35}_{-0{\cdot}15}$	$0{\cdot}64^{+0{\cdot}12}_{-0{\cdot}23}$
Hunts. Stibbington	5	$0{\cdot}26^{+0{\cdot}05}_{-0{\cdot}10}$	$0{\cdot}33^{+0{\cdot}07}_{-0{\cdot}06}$	$<0{\cdot}01^{+0{\cdot}00}$	$0{\cdot}92^{+0{\cdot}28}_{-0{\cdot}14}$	$0{\cdot}73^{+0{\cdot}08}_{-0{\cdot}10}$
Nr. Doncaster. Rossington Bridge	5	$0{\cdot}32^{+0{\cdot}11}_{-0{\cdot}07}$	$>1{\cdot}0$	$0{\cdot}035^{+0{\cdot}033}_{-0{\cdot}012}$	$0{\cdot}49^{+0{\cdot}11}_{-0{\cdot}23}$	$0{\cdot}24^{+0{\cdot}13}_{-0{\cdot}10}$
Cantley	3	$0{\cdot}33^{+0{\cdot}01}_{-0{\cdot}01}$	$1{\cdot}0^{+0{\cdot}15}_{-0{\cdot}18}$	$0{\cdot}016^{+0{\cdot}006}_{-0{\cdot}004}$	$0{\cdot}55^{+0{\cdot}21}_{-0{\cdot}17}$	$0{\cdot}31^{+0{\cdot}03}_{-0{\cdot}05}$
Cheshire. Wilderspool	5	$0{\cdot}59^{+0{\cdot}22}_{-0{\cdot}12}$	$>2_{-0{\cdot}0}$	$0{\cdot}061^{+0{\cdot}023}_{-0{\cdot}024}$	$0{\cdot}71^{+0{\cdot}24}_{-0{\cdot}16}$	$1{\cdot}16^{+0{\cdot}49}_{-0{\cdot}39}$
Warwickshire. Mancetter	11	$0{\cdot}33^{+0{\cdot}33}_{-0{\cdot}19}$	$>1{\cdot}0_{-0{\cdot}5}$	$0{\cdot}050^{+0{\cdot}034}_{0{\cdot}029}$	$1{\cdot}03^{+0{\cdot}37}_{-0{\cdot}38}$	$0{\cdot}31^{+0{\cdot}16}_{-0{\cdot}11}$
Hartshill	2	$0{\cdot}22^{+0{\cdot}05}_{-0{\cdot}02}$	$0{\cdot}70\pm0{\cdot}00$	$0{\cdot}011^{+0{\cdot}001}_{-0{\cdot}000}$	$0{\cdot}63^{+0{\cdot}09}_{-0{\cdot}08}$	$0{\cdot}41^{+0{\cdot}02}_{-0{\cdot}03}$
W. Midlands. Wroxeter area	10	$0{\cdot}21^{+0{\cdot}08}_{-0{\cdot}08}$	$0{\cdot}93^{+0{\cdot}27}_{-0{\cdot}21}$	$0{\cdot}017^{+0{\cdot}027}_{-0{\cdot}008}$	$0{\cdot}72^{+0{\cdot}17}_{-0{\cdot}25}$	$0{\cdot}32^{+0{\cdot}16}_{-0{\cdot}11}$
Herts/Middx. Group A	3	$0{\cdot}14^{+0{\cdot}02}_{-0{\cdot}02}$	$0{\cdot}52^{+0{\cdot}01}_{-0{\cdot}00}$	$<0{\cdot}01^{+0{\cdot}00}$	$0{\cdot}74^{+0{\cdot}18}_{-0{\cdot}14}$	$0{\cdot}60^{+0{\cdot}05}_{-0{\cdot}04}$
Group B	3	$0{\cdot}22^{+0{\cdot}04}_{-0{\cdot}05}$	$0{\cdot}47^{+0{\cdot}20}_{-0{\cdot}15}$	$0{\cdot}018^{+0{\cdot}006}_{-0{\cdot}003}$	$0{\cdot}99^{+0{\cdot}09}_{-0{\cdot}08}$	$0{\cdot}17^{+0{\cdot}03}_{-0{\cdot}03}$
Brockley Hill	7	$0{\cdot}15^{+0{\cdot}08}_{-0{\cdot}05}$	$0{\cdot}49^{+0{\cdot}25}_{-0{\cdot}18}$	$\sim0{\cdot}01$	$0{\cdot}37^{+0{\cdot}12}_{-0{\cdot}07}$	$0{\cdot}44^{+0{\cdot}16}_{-0{\cdot}12}$
Radlett	3	$0{\cdot}15^{+0{\cdot}03}_{-0{\cdot}02}$	$0{\cdot}44^{+0{\cdot}09}_{-0{\cdot}14}$	$<0{\cdot}01^{+0{\cdot}00}$	$0{\cdot}40^{+0{\cdot}09}_{-0{\cdot}14}$	$0{\cdot}34^{+0{\cdot}08}_{-0{\cdot}16}$
E. Anglia. Colchester (Essex)	7	$0{\cdot}35^{+0{\cdot}34}_{-0{\cdot}17}$	$0{\cdot}87^{+0{\cdot}43}_{-0{\cdot}22}$	$0{\cdot}038^{+0{\cdot}012}_{-0{\cdot}020}$	$0{\cdot}37^{+0{\cdot}11}_{-0{\cdot}11}$	$3{\cdot}4^{+1{\cdot}0}_{-1{\cdot}4}$
Hevingham (Norfolk)	2	$0{\cdot}32^{+0{\cdot}02}_{-0{\cdot}03}$	$0{\cdot}80^{+0{\cdot}20}_{-0{\cdot}20}$	$0{\cdot}86^{+0{\cdot}10}_{-0{\cdot}19}$	$0{\cdot}82\pm0{\cdot}02$	$1{\cdot}56^{+0{\cdot}06}_{-0{\cdot}06}$
Mon. Caerleon area	3	$0{\cdot}67\pm0{\cdot}02$	$1{\cdot}6\pm0{\cdot}3$	$0{\cdot}036^{+0{\cdot}030}_{-0{\cdot}020}$	$0{\cdot}43\pm0{\cdot}05$	$0{\cdot}26^{+0{\cdot}09}_{-0{\cdot}08}$
Dorset. Corfe Mullen	2	$0{\cdot}12^{+0{\cdot}02}_{-0{\cdot}03}$	$0{\cdot}26\pm0{\cdot}16$	$<0{\cdot}01+0{\cdot}00$	$0{\cdot}51^{+0{\cdot}11}_{-0{\cdot}12}$	$0{\cdot}18^{+0{\cdot}04}_{-0{\cdot}05}$

The percentages given are mean values, and the maximum divergences observed are indicated above and below this figure.

Table 2. CONCENTRATION RANGES FOR THE REFERENCE ELEMENTS IN COLOUR-COATED WARES REGARDED AS TYPICAL OF PARTICULAR SITES

Origin	No. of samples	Per cent Na_2O	Per cent MgO	Per cent MnO	Per cent TiO_2	Per cent CaO
Lincs. Swanpool	3	$0{\cdot}33^{+0{\cdot}06}_{-0{\cdot}05}$	$>1{\cdot}0$	$0{\cdot}046^{+0{\cdot}043}_{-0{\cdot}026}$	~5	$1{\cdot}1\pm0{\cdot}0$
Hunts. Chesterton	9	$0{\cdot}35^{+0{\cdot}08}_{-0{\cdot}08}$	$>1{\cdot}0$	$0{\cdot}013^{+0{\cdot}009}_{-0{\cdot}003}$	$1{\cdot}6^{+0{\cdot}8}_{-0{\cdot}8}$	$0{\cdot}82^{+0{\cdot}28}_{-0{\cdot}25}$
Essex. Colchester	5	$0{\cdot}48^{+0{\cdot}04}_{-0{\cdot}02}$	$>1{\cdot}0$	$0{\cdot}081^{+0{\cdot}011}_{-0{\cdot}016}$	$2{\cdot}6^{+0{\cdot}7}_{-0{\cdot}7}$	$\sim2{\cdot}5^{+0{\cdot}8}_{-0{\cdot}4}$
Rutland. Great Casterton	3	$0{\cdot}25^{+0{\cdot}04}_{-0{\cdot}04}$	$0{\cdot}57^{+0{\cdot}11}_{-0{\cdot}11}$	$<0{\cdot}01+0{\cdot}00$	$2{\cdot}6^{+0{\cdot}4}_{-0{\cdot}2}$	$1{\cdot}07^{+0{\cdot}23}_{-0{\cdot}11}$

The spectra were photographed in a Hilger large quartz spectrograph. 5 ± 0·05 mgm. of a 1 : 1 : 1 mixture of sample/graphite/ammonium sulphate containing 1·5 per cent lithium carbonate were arced at 8–9 amp. for 30 sec. in shallow graphite cups against a flat cathode. The arc gap was kept at 4 mm.; a 3-mm. diaphragm 2·5 cm. from it helped to reduce the background. The dilution of the sample was found to improve the burning properties of these refractory materials and to reduce matrix effects[2–4]. The lithium was also useful as an internal standard in some of the experiments.

Considerable systematic regional variations in the concentrations of calcium, sodium, titanium, magnesium and manganese were observed by visual comparison with the spectra of series of mixtures of oxides of 50 elements in an ammonium sulphate base arced under similar conditions. The variations in iron, copper, zirconium, vanadium, chromium and nickel content were either small or masked by the random fluctuations from sample to sample. By means of British chemical standard samples of silica brick and firebrick, calibration curves were set up for the first group of elements. To check the internal consistency of the method in the case of manganese, calcium, and titanium, calibration curves for two lines of different intensities were used. The concentrations of the chosen elements in typical samples were averaged for each kiln area and the results are shown in Table 1. Another 80–100 samples of less certain attribution on archæological grounds were then similarly analysed and the results compared with those in Table 1. In most cases the two kinds of evidence taken together have made the kiln area virtually certain. The details of this work will be published elsewhere, together with results for further kiln-site material as they become available.

From the results it seems that the number of elements for which the quantitative analyses were carried out is sufficient to differentiate between material from well-separated regions. The differences of concentration of the various elements from region to region, when taken together, should leave little ambiguity about the source of the material when deciding between a limited number of archæological alternatives. The decision is made easier in some cases: for example, the compositions of the samples from Lincoln show high titanium, and from Colchester high calcium.

The results for some colour-coated wares, selected by Mr. G. Webster (Table 2), also show significant trends. It is impossible to determine the origin of much of this pottery on purely archæological grounds, and it is for this kind of problem that the spectrographic method may become useful. The colour-coated samples have also been analysed for potassium, gallium, strontium, barium and lead by a different technique, and the results obtained so far suggest that these data will add further criteria of the origin.

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[1] Prelim. Reps. Reference Clay Minerals Amer. Pet. Inst., Research Project No. 49 (1951).

[2] Birks, F. T., Atomic Energy Research Establishment *C/R* 1177 (1953).

[3] Waring, C. L., and Annell, C. S., *Anal. Chem.*, **25**, 1174 (1953).

[4] Weaver, J. R., and Brattain, R. R., *Anal. Chem.*, **21**, 1038 (1949).

Neutron Activation Analysis of Ancient Roman Potsherds

THIRTY-TWO fragments of Roman samian ware have been analysed for their sodium and manganese content, using neutron activation and γ-ray spectrometry. They were chosen from at least 10 of about 30 factories at which this fine-grained pinkish-red Roman table-ware was manufactured. The body of the sherds from all factories is very similar in appearance and a chemical method was sought to differentiate between factories. Similar analyses on other types of pottery have been done by Sayre *et al.*[1,2] at Brookhaven National Laboratory, who found quantitative chemical differences in material from different places. Our results, summarized in Table 1, for 10 or more of the factories, show that this also holds in general for samian ware.

Table 1

Region	No. of factories	No. of sherds analysed	Manganese Average content*	Manganese Standard deviation	Sodium Average content*	Sodium Standard deviation
1. Italy	?	3	8	0·5	2·2	0·25
2. North-east Gaul (east of Moselle valley)	8	9	4	0 45	1·6	0·2
2 (*a*). Chémery, north-east Gaul (east of Moselle valley)	1	3	2	0·2	2·4	0·1
3. Central-south Gaul	3	11	3	0 15	0 7	0·15
4. North-east Gaul (west of Moselle valley)	2	6	3	0 1	0·6	0 1

* In arbitrary units.

50-mgm. samples from the interior of the sherds were irradiated together (so that results for different sherds would be comparable) for 4 hr. in a flux of $1 \cdot 1 \times 10^{12}$ neutrons/cm.^{2}/sec. in *Bepo* at Harwell. The quantities of the elements present were measured in terms of the intensity of the 0·84 MeV. manganese-56 γ-ray and the 2·76 sodium-24 γ-ray, and as no standards were used the results are in arbitrary units. The reactions considered were the thermal neutron reactions manganese-55 (n,γ) manganese-56 and sodium-23 (n,γ) sodium-24, any contribution to the total sodium-24 activity from the fast neutron reaction aluminium-27 (n,α) sodium-24 on aluminium in the sherds being negligible.

Sherds tended to group regionally, and one factory at Chémery in north-east Gaul (east of the Moselle valley) was distinct from the other three in the region. That manufactured close at hand and to the west of this valley is also distinct (group 4), but on the other hand, does not differ significantly from that made in south and central Gaul. Whatever the reason for these differences they are of archæological interest, and suggest a means for locating the region of origin of samian sherds which cannot be placed stylistically.

We gratefully acknowledge the help of Mrs. E. Heckmann, who initiated the work and developed the gamma-ray spectrometer used.

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[1] Sayre, E. V., and Dodson, R. W., *Amer. J. Archaeol.*, **61**, 35 (1957).

[2] Sayre, E. V., Murrenhoff, A., and Weick, C. F., Brookhaven National Laboratory, Chemistry-General, **122**, 508 (1958).

FORTHCOMING EVENTS

*(Meetings marked with an asterisk * are open to the public)*

Monday, January 18

Royal Geographical Society (at 1 Kensington Gore, London, S W.7), at 5 p.m.—Mr. Peter Haggett: "Landuse in the Serra do Mar, Brazil".

University of London (in the Meyerstein Lecture Theatre, Westminster Medical School, Horseferry Road, London, S.W.1), at 5.15 p.m.—Prof. V. M. Trikojus (University of Melbourne): "Some Aspects of Thyroid Biochemistry".*

British Society for the Philosophy of Science (in the Eugenics Theatre, University College, Gower Street, London, W.C.1), at 5.30 p.m—Prof. K. R Popper "An Indeterminist's View of the Physical World" (Presidential Address).

Society of Chemical Industry, Pesticides Group (at 14 Belgrave Square, London, S W.1), at 5 30 p.m.—Dr. L. Crombie: "Lipid *Iso*butylamides".

Royal Institute of Chemistry (at Enfield Technical College, Queensway, Ponders End, Enfield, Middlesex), at 7.30 p.m.—Dr D K. Hill. "Recent Advances in Glass Technology".

Monday, January 18—Friday, January 22

Physical Society (at the Royal Horticultural Society's Halls, Westminster, London, S.W.1)—44th Exhibition of Scientific Instruments and Apparatus.

Tuesday, January 19

University College (in the Anatomy Theatre, Gower Street, London, W.C.1), at 1.15 p.m.—Dr. A. Comfort: "Darwin and Freud".*

Institution of Electrical Engineers, Measurement and Control Section (at Savoy Place, London, W C.2), at 5.30 p m.—Discussion on "Superseding the Ratio Arms in A.C. Bridges", opened by Mr. C. G. Mayo.

University College (in the Physics Theatre, Gower Street, London, W.C 1), at 5.30 p m.—Prof. Alonzo Church (Princeton University): "Logic and Meaning".* (First of four Shearman Memorial Lectures. Further Lectures on January 21, 26 and 28.)

Plastics Institute (at the Wellcome Building, 183–193 Euston Road, London, N.W.1) at 6.30 p.m.—Dr. G. Swift: "Plastics in the Packaging Industry".

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Dr. J. Seddon: "Some Aerodynamic Problems of Engine Installation".

Pharmaceutical Society of Great Britain (at 17 Bloomsbury Square, London, W.C.1), at 7.30 p.m.—Dr N. J. Harper. "A Three Dimensional Approach to Drug Design".

Wednesday, January 20

Physical Society, Acoustics Group (in the Physics Department, Imperial College, Imperial Institute Road, London, S.W.7), at 2.30 p.m.—Symposium on "Vibrations".

Royal Meteorological Society (at 49 Cromwell Road, London, S.W.7), at 5 p m.—Monsieur N. Gerbier (Paris): "Experimental Studies of Wave-form Clouds"; Dr. E. Palm (Oslo): "Recent Developments in Theoretical Treatment of Mountain Waves".

Royal Statistical Society (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.15 p.m.—Mr. K. F. Glover: "Statistics of the Transport of Goods by Road".

Eugenics Society (at the Royal Society, Burlington House, Piccadilly, London, W.1), at 5.30 p.m—Mr. Robert H. S. Robertson: "The Output of Scientists in Scotland—1600 to 1950".

Institute of Physics (at 47 Belgrave Square, London, S.W.1), at 6 p.m.—Mr. H. J. Dunster: "Health Physics".

Institution of Electrical Engineers, Education Discussion Circle (at Savoy Place, London, W.C.2), at 6 p m.—Discussion on "One Approach to Generalized Machine Analysis", opened by Mr. G. J. Henderson.

Royal Institute of Chemistry (joint meeting with the British Society for the History of Science, at the Science Museum, Exhibition Road, South Kensington, London, S.W.7), at 6.30 p.m.—Mr F. Greenaway: "Thirty Centuries of Assaying".

Oil and Colour Chemists' Association, London Section (at the Royal Society of Tropical Medicine and Hygiene, Manson House, 26 Portland Place, London, W.1), at 7 p m.—Mr. G. L. Cooper: "An Appreciation of Work Study", followed by a film "Introducing Work Study".

Society for Analytical Chemistry, Biological Methods Group (at the Chemical Society, Burlington House, Piccadilly, London, W.1), at 7 p.m.—Prof. L. Garrod: "Antibiotic Assays in Body Fluids".

Society for Psychical Research (at Kensington Town Hall, Kensington High Street, London, W.8), at 7 p.m. Dr. S. G. Soal: "The Jones Boys Experiments—The Case Against Cheating".

Wednesday, January 20—Thursday, January 21

Institution of Electrical Engineers, Measurement and Control Section (in co-operation with the British Computer Society, at Savoy Place, London, W.C.2)—Discussion Meetings on "Reliability and Maintenance of Digital Computer Systems—Managerial and Engineering Aspects".

Thursday, January 21

Institution of Mining and Metallurgy (at the Geological Society, Burlington House, Piccadilly, London, W.1), at 5 p.m—Mr. J. H. Harris "Serial Gravity Concentration—a New Tool in Mineral Processing"; Mr. P. D. R. Maltby "Use of Moving Bed Ion Exchange in the Recovery of Uranium at Can-Met Explorations, Ltd., Blind River, Ontario".

London Mathematical Society (at the Royal Astronomical Society, Burlington House, Piccadilly, London, W.1), at 5 p.m—Dr. S. J. Taylor: "Properties of the Pólya Random Walk".

Institute of Information Scientists (at Caxton Hall, Caxton Street, London, S.W.1), at 6 p.m.—Mr. R. L. Brown: "The Critical Technical Review and Its Contribution to Progress".

Institution of Mechanical Engineers, Hydraulics Group (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Discussion on "The Value of Hydraulics Research in Mechanical Engineering"

Society of Chemical Industry, Road and Building Materials Group (at 14 Belgrave Square, London, S.W.1), at 6 p.m.—Mr. D. B. Honeyborne "Plastics for Building".

Royal Society of Tropical Medicine and Hygiene (at Manson House, 26 Portland Place, London, W 1), at 7.30 p.m.—Symposium on "Arthropod-borne Diseases"; Dr. C. E. Gordon Smith: "Factors in the Past and Future Evolution of the Arboviruses"; Mr. P. F. Mattingly. "Ecological Aspects of the Evolution of Mosquito-borne Virus Diseases"; Prof. C. D. Darlington "Origin and Evolution of Viruses".

Saturday, January 23

Biochemical Society (in the Department of Biochemistry, London Hospital Medical College, Turner Street, London, E.1), at 10 a.m—390th Meeting

London County Council (at the Horniman Museum, London Road, Forest Hill, London, S.E 23), at 3.30 p.m.—Mr. Philip Sumner: "The Wheel in the History of Transport".*

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned

Assistant Lecturer in the Department of Sociology—The Registrar, The University, Leicester (January 30).

Chief Cataloguer (graduate with good cataloguing experience, preferably of the Library of Congress scheme)—The Registrar, The University, Nottingham (January 30).

Lecturer (honours graduate in physics or chemistry) in Polymer Chemistry in the Department of Textile Chemistry—The Registrar, The Manchester College of Science and Technology, Manchester 1 (January 30).

Lecturer in Physics and/or Chemistry—The Principal, Borough Road College, Isleworth, Middlesex (January 30).

Research Assistant in the Department of Pure Mathematics—The Registrar, The University, Liverpool, quoting Ref CV/T (January 30)

Scientific Officer (male, with an honours degree), for genetical and cytological research—The Secretary, Plant Breeding Institute, Trumpington, Cambridge (January 30).

Assistant Lecturer in Biochemistry—The Registrar, The University, Sheffield (January 31).

Boots Medical Research Fellow (medically qualified), to undertake research in one of the following fields: Pharmacology, Therapeutics, Pathology, Bacteriology, Medicine, Surgery, or Obstetrics and Gynæcology—The Registrar, The University, Sheffield (January 31).

Lecturer in Bacteriology—The Registrar, The University, Sheffield (January 31).

Assistant Lecturer in Biochemistry—The Registrar, The University, Sheffield (January 31).

Assistant Lecturer in Mathematics—The Registrar, University College of North Staffordshire, Keele, Staffordshire (February 1).

Lecturer/Senior Lecturer in Mechanical Engineering at the University of Sydney, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, February 2).

Senior Lecturer or Lecturer, and a Lecturer in the Department of Mechanical Engineering, University of Western Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, February 8)

Chair of Dermatology at the Institute of Dermatology—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 10).

Reader in Laryngology at the Institute of Laryngology and Otology—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 11).

Experimental Officer/Assistant Experimental Officer (electronic engineer with experience of pulse techniques) at the National Physical Laboratory (Metallurgy Division), Teddington, Middlesex, for the upkeep and development of the following equipment: nucleonic counters, X-ray detectors, spectrographic source units, automatic timers, voltage regulators, mass spectrometers, and wide band amplifiers—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. D.633/9A (February 12).

LECTURER IN PHYSICS, and a LABORATORY SUPERINTENDENT IN THE DEPARTMENT OF PHYSICS, Fourah Bay College, Sierra Leone (University of Durham)—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (February 12).

ASSISTANT LECTURER or LECTURER (with a degree in pure science) IN AGRICULTURAL ZOOLOGY at Canterbury Agricultural College, University of New Zealand—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C 1 (New Zealand and London, February 15).

CHAIR OF ORGANIC CHEMISTRY at Queen Mary College—The Academic Registrar, University of London, Senate House, London, W C 1 (February 15).

SENIOR LECTURER and a LECTURER IN PHYSICS at Victoria University of Wellington, New Zealand—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (New Zealand and London, February 29).

SENIOR LECTURER IN AGRICULTURAL ECONOMICS at Canterbury Agricultural College, University of New Zealand—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W C.1 (New Zealand and London, February 29).

SENIOR LECTURER or LECTURER IN EDUCATION at the University of Otago, Dunedin, New Zealand—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1, or from the Registrar of the University (New Zealand and London, February 29).

ASSISTANT TUTOR or TUTOR IN PHARMACEUTICS; an ASSISTANT TUTOR or TUTOR IN PHARMACEUTICAL CHEMISTRY; an ASSISTANT TUTOR or TUTOR IN PHARMACOGNOSY; and an ASSISTANT TUTOR IN PHARMACOLOGY at the New Zealand School of Pharmacy—The High Commissioner for New Zealand, 415 Strand, London, W.C.2 (March 1).

BIOCHEMIST or MICROBIOLOGIST (with an honours degree), to work in the field of bacterial metabolism—The Secretary, Medical Research Council of Ireland, 50 Merrion Square, Dublin (March 1).

PETER NICOL RUSSELL CHAIR OF MECHANICAL ENGINEERING at the University of Sydney, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, March 15).

ASSISTANT PHYSICIST (with an honours degree in physics and postgraduate experience in radiation or nuclear physics) IN THE DEPARTMENT OF PHYSICS APPLIED TO MEDICINE, for duties concerned mainly with assisting in the organization of practical work, demonstrations and teaching aids for postgraduate courses for the M.Sc. degree in Radiation Physics, and other postgraduate courses in radiological physics and protection—The Secretary, Department of Physics Applied to Medicine, Middlesex Hospital Medical School, London, W.1.

ASSOCIATE PROFESSOR or ASSISTANT PROFESSOR (theoretical physicist, preferably with experience and main interest in the field of nuclear theory), to teach courses at the senior and postgraduate levels and to carry on research—The Head, Department of Mathematical Physics, University of Manitoba, Winnipeg, Canada.

BIOLOGIST (with a good degree in physiology or zoology and considerable research experience, preferably with spermatozoa); and a BIOCHEMIST (with a good degree in biochemistry or chemistry, and research experience with polysaccharides or proteins, preferably involving work with animals), to join a small research team investigating cervical mucus, with particular reference to problems of fertility —The Secretary, National Institute for Research in Dairying (University of Reading), Shinfield, Reading, quoting Ref. 59/17.

BIOLOGIST (with a strong leaning towards botany, and capable of teaching biology to *A* level and botany to *S* level); and a CHEMIST (capable of teaching the subject to *A* level)—The Head Master, Westminster School, Dean's Yard, London, S.W.1.

CHIEF TECHNICIAN (trained in hæmatology, able to supervise staff and plan systematic experimental work)—The Personnel Manager, Jewish General Hospital, 3755 Cote Street, Catherine Road, Montreal, Quebec, Canada.

GRADUATE ASSISTANTS (qualified to pursue M.Sc. and Ph.D. work) IN CHEMISTRY—The Chairman, Department of Chemistry, McMaster University, Hamilton, Ontario, Canada.

INDEXER (female, preferably graduate)—The Director, Commonwealth Bureau of Pastures and Field Crops, Hurley, near Maidenhead, Berks

INSTRUCTOR or ASSISTANT PROFESSOR; POSTDOCTORATE FELLOWS; and SCHOLARS IN THE DEPARTMENT OF PHYSICS—The Head, Department of Physics, University of British Columbia, Vancouver, B.C., Canada.

ISOTOPE TECHNICIAN (preferably with a knowledge of biochemistry or electronics), for a specialized laboratory dealing with the diagnosis, treatment and research in thyroid disorders—Hospital Secretary, New End Hospital, Hampstead, London, N.W.3.

LABORATORY TECHNICIAN (A.I M.L T. or B.Sc.), for routine hospital biochemistry—The Secretary, Group 9 Pathological Laboratory, 6 Rickmansworth Road, Watford.

LECTURER (with an honours degree in mathematics or statistics, with teaching and research or industrial experience involving the application of mathematics to scientific and engineering problems) IN MATHEMATICS—The Registrar, College of Technology, Ashley Down, Bristol 7.

PROFESSOR'S SECRETARY (preferably graduate, especially botany or horticulture), to work at the University of Reading Horticultural Research Laboratories—The Assistant Bursar, The University, Reading.

RESEARCH ASSISTANT (with some previous laboratory or animal experience), for work on the biochemistry of the eye—The Secretary, University College Hospital Medical School, University Street, London, W.C.1.

RESEARCH FELLOW IN THE DEPARTMENT OF MEDICAL PHYSICS, for work concerned with the physical problems arising from the use of ionizing radiations in medicine—The Secretary, Royal Free Hospital School of Medicine (University of London), 8 Hunter Street, London, W.C.1.

SCIENTIFIC OFFICER (with a first- or second-class honours degree in physics or equivalent) at the National Physical Laboratory (Standards Division), Teddington, Middlesex, to apply modern techniques, for example, optical interference, to precision measurement in first place of quantities, such as length and pressure, and later of fundamental physical quantities and constants—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting A.577/9A.

SENIOR PHARMACIST at the London Chest Hospital—The House Governor, London Chest Hospital, London, E.2.

TECHNICAL ASSISTANT (preferably with G.C.E. "A" level, or better, in physics, chemistry and biology), for research in cell structure and nucleic acids, using the electron microscope, etc.—The Director, M.R C. Biophysics Research Unit, King's College, Strand, London, W.C.2.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

London and Home Counties Regional Advisory Council for Technological Education. Engineering Education in the Region. Seventh edition. Pp. 36. (London: London and Home Counties Regional Advisory Council for Technological Education, 1959.) 3*s* 6*d*. [3011

Ciba (A.R.L.), Ltd. Technical Notes, No. 208 (November, 1959): 'Redux' for Bonding Brake Liners. Pp. 8. (Duxford: Ciba (A.R.L.), Ltd., 1959.) [3011

Committee on Science and Freedom. Bulletin No. 13: The Scholar and Society—Report on a Study Group held in Tunis, April, 1959. Pp. 144. (Manchester. Committee on Science and Freedom, 818, Wilmslow Road, 1959.) [3011

Department of Scientific and Industrial Research. Automatic Control in Soviet Industry: a Report of a visit to the Soviet Union in May, 1959. Pp. 64. (London: Department of Scientific and Industrial Research, 1959.) [3011

The British Rubber Producers' Research Association. List of Publications, 1940–1959, and Supplements. Pp. 32. (London: British Rubber Producers' Research Association, 1959.) [3011

Building Research Station Digest No. 127 (October, 1959): Simplified Plumbing for Multi-storey Flats. Pp. 4. (London · H.M. Stationery Office, 1959.) 4*d*. [3011

Other Countries

World Health Organization. Library News, Vol. 12, Supplement No. 2: Current Indexing and Abstracting Periodicals in the Medical and Biological Sciences—an Annotated List. Second edition. Pp. 85. (Geneva World Health Organization, 1959.) [3011

Académie Royale de Belgique. Classe des Sciences. Mémoires, Tome 31, Fascicule 6: Flexion et Extension des Plaques d'Epaisseur Modérée. Par Prof. B. Fraeijs de Veubeke. (Bruxelles: Académie Royale de Belgique, 1959.) [3011

East Africa High Commission. East African Agriculture and Forestry Research Organization. Record of Research for the period January 1 to December 31, 1958—Annual Report 1958. Pp. vii+109. (Nairobi · Government Printer, 1959.) Shs. 3/50. [3011

United States (86th Congress, 1st Session). United States Foreign Policy. Possible Nonmilitary Scientific Developments and their Potential Impact on Foreign Policy Problems of the United States: a Study prepared at the request of the Committee on Foreign Relations, United States Senate, by Stanford Research Institute. Pp. ix+100. (Washington, D C.: Government Printing Office, 1959.) [3011

The Regional Research Centre of the British Caribbean at The Imperial College of Tropical Agriculture, Trinidad, W.I. Soil and Land-Use Surveys No. 6: British Guiana. 1 · The Rupununi Savannas (continued). 2: The Intermediate Savannas. 3 · General Remarks. By J. Stark, G. K. Rutherford, J. Spector and T. A. Jones. Pp. 24. (St. Augustine, Trinidad: Imperial College of Tropical Agriculture, 1959) 15*s*. [3011

Universidade de São Paulo. Boletim do Instituto Oceanografico, Tomo VIII, Fasc 1 e 2. Pp. 261. Contribuicoes Avulsas do Instituto Oceanografico. No. 1, 1957 *Cokeridia kathetostoma* n. sp., da fam. *Dactyloscopidae* (Pisces-Perciformes) By J. de Paiva Carvalho. Pp. 5+1 plate. No. 2, 1957: Istituto de Biologia Maritima das Caraibas · Fundacao e Patrocinador. Pp 5. No. 1, 1958: Red para Fitoplancton con copo Intercambiable. Pp. 5+2 plates. (São Paulo: Universidade de São Paulo, Instituto Oceanografico, 1957 and 1958.) [3011

Instituto Ecuatoriano de Ciencias Naturales, Quito, Ecuador. Contribucion No. 29 Los Manglares del Ecuador. Por Dr. M. Acosta-Solis. Pp. 82. (Quito: Instituto Ecuatoriano de Ciencias Naturales, 1959.) [3011

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CONTROL OF RADIOACTIVE WASTES

THE recent Monaco Conference on Radioactive Waste Disposal (see *Nature* of January 2, p. 1) was held at about the same time as a Bill dealing with the subject was introduced in the House of Lords. At the present time, disposal of radioactive waste in Great Britain, by organizations other than the United Kingdom Atomic Energy Authority, is governed by the same regulations as apply to ordinary domestic and industrial wastes. With the growing use of radioactive substances by a diversity of bodies and because of certain hazards peculiar to the disposal of radioactive wastes, it was realized that these regulations were probably inadequate. In 1956, at the invitation of the Minister of Housing and Local Government, an expert panel was appointed by the Radioactive Substances Committee with the following terms of reference :

"To ascertain the nature and quantity of radioactive waste likely to arise in the foreseeable future ; to advise on the best methods of securing that the waste is disposed of safely ; and to advise whether any new legal provisions or amendments to existing legislation are necessary to ensure safe disposal, and, if so, to advise on the form which the new provisions or amendments should take."

The report of this Committee is contained in a recent White Paper* and the recommendations of the Committee are effectively incorporated in the Radioactive Substances Bill, introduced in the House of Lords on November 13. The Bill provides that the temporary powers by which the discharge of radioactive wastes by the Atomic Energy Authority and other licensed nuclear installations are exercised by the Minister of Housing and Local Government and the Minister of Agriculture, Fisheries and Food, and in Scotland the Secretary of State, will be made permanent. Other users of radioactive materials will be obliged to register their premises with the appropriate Minister (in England and Wales, the Minister of Housing and Local Government and in Scotland, the Minister of State). No disposal of radioactive waste will be permitted except on authorization from the appropriate Minister or Ministers. The Ministry of Housing and Local Government and the Secretary of State for Scotland will, in addition, have power to arrange a national disposal service to handle wastes that cannot safely be dealt with in the area in which they originate. The cost of this service will be chargeable to users of the service.

Both the principles and recommendations emerging from the report of the Committee should be studied by all present and future users of radioactive substances. The views expressed may be briefly summarized. There must be adequate justification for the use of any source of ionizing radiation, no matter how small. Subject to such justification and with suitable precautions, the use of radionuclides should be encouraged. It is not possible to dispose of radioactive wastes without adding to the background irradiation to which the public is exposed, but it is possible to keep this increase well within acceptable limits. Irrespective of cost, it must be ensured that the discharge of radioactive waste does not endanger the health of any individual member of the public, and further, in the terms of the White Paper, "in considering whether a given discharge should or should not be permitted, it is necessary to decide whether the radiation from the discharge, when added to that from all other sources of radiation, is likely to result in a genetic hazard to the nation as a whole ; from this point of view it is as much the concern of, for example, London as of Manchester if the inhabitants of either are exposed to ionizing radiations". It is obvious that local control of discharges would not enable such decisions to be taken, and that a national body staffed by suitable experts would be necessary. When considering the level of permissible discharge, irradiation of the public by other sources, notably normal background, X-ray and fall-out, has to be considered. The Committee suggests that the gonad irradiation arising directly from waste disposal, to which the whole population is exposed, should in no circumstances be permitted to exceed an average dose of 1 r. per person per 30 years, and also emphasizes that it would expect that, with reasonable precautions, the average dose would be maintained much below this level. The recommendation of the Committee for the setting-up of a National Disposal Service in no way suggests that local disposal is unsatisfactory ; on the contrary, local disposal, wherever feasible, is recommended. It is anticipated that the primary function of the National Disposal Service will be to dispose of highly active wastes of long half-life.

A detailed and quantitative picture of the present scale and range of radioactive waste disposal emerges from the report of the Committee. It would appear that, with a few notable exceptions, particularly in the luminizing industry, the hitherto limited controls have so far ensured that radioactive wastes have been disposed of in an acceptable manner. The Committee has been in much greater difficulty, however, when attempting to forecast the quantity of radioactivity to be disposed of in the future. Thus, when considering wastes likely to arise from private reactors, its comment that "we find it difficult to estimate the magnitude of the wastes likely to be produced in these enterprises", emphasizes the need for an organization responsible for radioactive waste disposal on a national basis and for the frequent review of limits and methods of disposal.

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* The Control of Radioactive Wastes. Pp. 46. (Cmnd. 884.) (London : H.M. Stationery Office, 1959.) 2*s*. 6*d*. net.

The Committee was in obvious difficulty in suggesting a limit for the irradiation dosage that could be received by the population as a result of waste disposal, in that "there is no agreed permissible genetic dose and there is no agreed allocation of dose to the different ways in which the population is irradiated". Whatever the research effort, it is difficult to imagine a quick answer to this problem. For immediate use, it would appear reasonable to suggest limits, as the Committee has done, which are considerably less than those which informed medical opinion regards as maximal. The report also indicates the need for considerable research in a number of other fields where information is lacking. Thus, when speaking of the discharge of liquid wastes to rivers, the Committee comments that "we know all too little of the absorption on river muds, the absorption into weeds and the take-up by cress". A research inquiry would appear to be required to establish the fate of radionuclides disposed of in sewage systems. It should not be impossible to ascertain what proportion of radionuclides, particularly those such as radiostrontium which are taken up by plants, pass through sewage disposal works and what proportion is retained in the sewage sludge, which may be used as a fertilizer. The report warns of the need for further research before highly active fission products produced as waste from the processing of fuel elements be dumped into the sea. The repeated reference to the lack of experts with the knowledge to deal with the problems of waste disposal poses the question as to whether adequate steps have been taken to ensure the training of sufficient personnel for the efficient working of the proposed central authority.

GEOPHYSICS FOR THE ENGINEER

Geophysical Surveys in Mining, Hydrological and Engineering Projects, 1958

Pp. viii+270. (The Hague, 30 Carel V. Bylandtlaan: European Association of Exploration Geophysicists, 1959.) 16.50 florins; 4.50 dollars.

THOSE who have followed the growth of geophysics since the early 'twenties will remark the happy circumstance that has allowed the publication of this excellent volume of case-histories to occur during Prof. J. M. Bruckshaw's term as president of the European Association of Exploration Geophysicists. Prof. Bruckshaw was a member of the original team which, under Broughton-Edge, produced one of the classics of applied geophysics, based on tests of all the different physical tools against a series of geological structures in Australia.

When it became apparent that geophysical methods could locate potential oil reservoirs, the oil industry application swamped all others. There has been a tendency, as with oil-well drilling, for the tools to be elaborated instrumentally, but, as Prof. Bruckshaw pointed out in his presidential address to the European Association of Exploration Geophysicists, there have been no fundamental changes in the methods that were developed thirty years ago. It is possible that present geophysical methods in the oil industry have already been pushed beyond their economic limits, and it may be opportune to take another look at how the mining and civil engineers use geophysics.

The engineer, whether looking for ore-bodies, locating water-supplies or studying the foundations of buildings or dam sites, cannot afford the luxury of rapid coverage of large areas with routine magnetic, gravity and seismic measurements. He must find the most suitable tool or combination of tools—including the drill—to solve his geological problem. In one respect, of course, the technical problems of the engineer are simplified, since all his underground structures are much shallower than most oil reservoirs, and often cheap and rapid electrical prospecting methods are adequate. However, the case-histories do show a proper appreciation of how geophysical tools should be used in geological work, as, for example, in some of the Rhône valley investigations, where electrical surveys were supplemented by seismic refraction measurements.

It is instructive to the oil geophysicist to read of the wide variety of successful applications of geophysical methods. In Yugoslavia, for example, ancient Roman mining activity is being extended by the exploration of a whole zone of electrical anomalies. In Finland, swamp-covered ore deposits are revealed by magnetic and electromagnetic surveys. The search for uranium-vanadium ores in Colorado in 1940 led to the use of the Geiger counter as a new prospecting tool It is pleasant also for those who have spent a great deal of time listening patiently to the claims of water diviners to see that great success has been achieved in the State of Illinois by using resistivity surveys to locate comparatively small pockets of water-bearing sand and gravel. The drill has shown that in hundreds of surveys the interpretations are 90 per cent correct, a better result than can be obtained by divining.

This collection of past experience should prove of great benefit to civil engineers, since there are many cases, such as the proposed Channel Tunnel or the new Forth Bridge, where the application of geophysical methods can supplement and extrapolate the information obtained traditionally by boring.

T. F. GASKELL

ELEMENTARY PARTICLES

The Theory of Elementary Particles

By Dr. J. Hamilton. (International Series of Monographs on Physics.) Pp. xii+482. (Oxford: Clarendon Press; London: Oxford University Press, 1959.) 75*s*. net.

IT must be said immediately that this is not a narrative book that aims to introduce one to elementary particles or indeed to interest one in them by persuasion at all. It is a book for the professional which deals with the mathematical formalism and techniques that have been found useful for our description of elementary particles and their interactions. The particles themselves, of course, figure and are the subject of a table at its end. They are used, however, rather more to illustrate the formalism than as the subjects of the work itself. The formalism presented is 'useful' in the sense that it is of immediate application in our everyday handling of the elementary particles. There is very little attempt to discuss mathematics in a quasi-philosophical framework as a source of concepts. In other words, this is a book that tells you how to do the standard calculations about elementary particles using the techniques that have been evolved since the War. It is therefore an extremely useful book and will be of the greatest practical value, particularly as a very thorough and general introduction for theoretical students. It is a book for which a great need has been felt for many years, and it satisfies that need. The writing is of considerable clarity, and it will not be merely the theoretical student who profits by the use of this work. The experimentalist who does not mind a little hard work is well able from this text to put himself in contact with the 'right' way of thinking about particles and their interactions. Helpful examples abound and the author has to be thanked for the large number of 'hints' that he gives in liberal footnotes to sustain the weaker brethren. In short, it is a technical work that will satisfy the professional theoretician without repelling the curious experimentalist. The author is to be both congratulated and thanked.

The early chapters describe the setting up of classical fields and lay stress on general invariance properties of a relativistic origin. Then follow discussions of the quantization of boson and fermion fields and the interaction of fields. This first half of the book is essentially introductory in character and sets out the formalism at some length in a clear and agreeable way. Renormalization methods are then introduced with a discussion of the S-matrix and scattering theory, followed by Heisenberg and interaction representation operators. The Chew-Low theory is discussed in some detail, and there is a good treatment of dispersion relations. Throughout the book the author rightly stresses the relation between the invariance properties and selection rules, and these are discussed at some length. There are final chapters on polarization analysis and the foundations of field quantization.

This is basically a book to be learnt from and to be used rather than one purporting to summarize the state of play. It will be widely appreciated.

D. H. WILKINSON

PETROLEUM INDUSTRY

Advances in Petroleum Chemistry and Refining

Vol. 1. Pp. xv+641. Vol. 2. Pp. xviii+634. Edited by Kenneth A. Kobe and John J. McKetta, Jr. (New York: Interscience Publishers, Inc.; London: Interscience Publishers, Ltd., 1959.) 109*s*. per volume.

THE petroleum industry has always maintained large and active research and development organizations, the contribution of which to the advancement of science is published in patent or technical literature in ever-increasing quantity. The annual reviews of "Petroleum Technology", published by the Institute of Petroleum, permitted a rapid appraisal of the state of knowledge in any particular aspect of the industry, but the increasing quantity of technical publications which had to be reviewed caused the editors to cease this most useful publication in 1952. No adequate replacement has yet appeared. Certain specific aspects of the subject have been collected in such works as the "Physical Chemistry of Hydrocarbons", edited by A. Farkas (1950), and the "Chemistry of Petroleum Hydrocarbons", edited by B. T. Brooks, C. E. Boord, S. S. Kurtz and L. Schmerling (1954). These contain chapters by a number of authors dealing with the state of knowledge of a particular aspect of the chemistry of hydrocarbons, but there is apparently no intention of publishing additional volumes which might serve to bring the subject-matter up to date.

A real need exists for this purpose, which has been recognized by the editors of the new publication, "Advances in Petroleum Chemistry and Refining". The editors state that the various aspects of the petroleum industry, economics and future trends, unit operations and design, refinery processes, petrochemicals and mechanical equipment, will be dealt with by specialist authors in separate chapters of a series of volumes which "over the course of several years will assume the character of a reference book". The two volumes under review constitute the first of this series, and contain a number of chapters devoted to each of the five group headings outlined previously. The individual authors are all active in research or development within the petroleum industry and many are already recognized as outstanding authorities on their subject.

The task of the individual authors has been simplified in that they are not bound by any restriction concerning the years to which their review must be devoted, nor apparently is it considered necessary to include a complete bibliography on the subject at the end of each chapter. Each author makes a critical appraisal of the state of knowledge in his particular subject at the time of writing and illustrates his comments by reference to such published work as he considers necessary for his purpose. In some instances, such as the excellent chapters on "Solvent Refining" (A. W. Francis and W. H. King) and on "Solid Polymers from Surface Catalysts" (H. N. Friedlander and W. Resnick), a fairly complete bibliography has been given, but in general the editors have not duplicated the bibliographies published annually in the Unit Process and Unit Operations review of *Industrial and Engineering Chemistry*. This permits a certain leisurely approach to the subject; the authors do not feel they have to cram a number of original references into every sentence but can deal at some length with the fewer references they choose to examine. The result is what our American friends might describe as an 'all time high' in technical review literature. The chapters are readable, and although not all of the same high standard are nevertheless interesting and in many cases stimulating.

Chemists and technologists in the petroleum industry will appreciate the critical treatment of the technical and patent literature by several of the authors, while the non-specialist will find much of general interest, particularly in the section dealing with economics and future trends. It is not often that books of this nature, intended for a specialized section of industry, can be recommended for general student reading, but in this case the books can be recommended to degree students and research workers in chemistry or chemical engineering. In particular, the chapter on "Solid Polymers from Surface Catalysts", by H. N. Friedlander and W. Resnick, is an excellent account of a rapidly developing field of chemistry which as yet has not been described in current text-books. Those interested in industrial developments will find the chapter dealing with "Raw Materials for the Petrochemical Industry", by P. W. Cornell and V. N. Hurd, of particular interest.

If the high standard of the first two volumes is maintained in future publications the series will prove a most welcome addition to the libraries of petroleum technologists, but it will only be welcome on these already overburdened shelves if it maintains its selective character and does not become a reference book.

F. MORTON

CHEMICAL AND BIOLOGICAL ACTION OF RADIATIONS

Radiolyse de Liquides Organiques

Par Milton Burton. Polymérisations Amorcées par les Radiations Ionisantes. Par A. Chapiro et M. Magat. Effets des Rayonnements de Grande Énergie sur les Polymères. Par A. Charlesby. (Actions Chimiques et Biologiques des Radiations, Troisième Série. Collection dirigée par M. Haissinsky.) Pp. iv+222. (Paris: Masson et Cie., 1958.) Broché, 4,000 francs; cartonné toile, 4,800 francs.

Les Peroxydes Organiques en Radiobiologie

(Organic Peroxides in Radiobiology). Par R. Latarjet *et al.* (Actions Chimiques et Biologiques des Radiations, Quatrième Série. Collection dirigée par M. Haissinsky.) Pp. viii+156. (Paris: Masson et Cie., 1958.) Broché, 2,800 francs; cartonné toile, 3,600 francs.

IN the third volume of Haissinsky's series of monographs the dominant theme, in each of the three topics discussed, is a physicochemical interpretation of the principal radiation effects. The various authors give at the outset a compact account of the theoretical principles which are used in the interpretation, and, in consequence, the articles serve as useful introductions to complex and expanding subjects. A further advantage of the approach is to focus attention on the studies which have led to results of a definitive character. It becomes apparent from the discussions that a great deal of published work in this field is of limited value, due to the fact that inadequate attention has been given to experimental conditions, such as purity of materials, methods of irradiation, and so on.

The theoretical background presented in the article by Burton is to some extent covered by Gray in the first volume of this series, but the reiteration, with greater emphasis on the role of excitation processes, and attention throughout to photochemical analogy, seems appropriate to the purpose in hand. His account of the radiolytic effects includes a range of organic liquids in various chemical classes, although the results obtained with hydrocarbons, which are the most susceptible to interpretation, occupy the major part of the discussion. The other two articles are authoritative and notably lucid. They will no doubt be read by radiobiologists in search of clues, but they also merit the attention of industrial polymer chemists.

The fourth volume is devoted to the proceedings of a colloquium concerned with an evaluation of the concept of organic peroxides as causal agents in radiobiological damage. Sixteen papers are presented, commencing with a useful account of recent work on the nature of the peroxides resulting from the autoxidation of fats. (The absence of literature references to this paper is an unfortunate omission.) The other topics range from the radiochemical production of peroxides in *in vitro* systems (cumene, polystyrene, nucleic acids), to the biological effects of peroxides and the influence of oxygen on radiation effect produced in a number of biological systems. Th discussions on the individual papers, and a valuabl discussion on the main theme of the colloquium, ar freely recorded in this volume.

The high standards set in the first two volume of this series are well maintained in the presen additions..

W. ANDERSON

SCIENTIFIC MEDICINE FOR POSTGRADUATES

Lectures on the Scientific Basis of Medicine

Vol. 7, 1957–58. (British Postgraduate Medica Federation: University of London.) Pp. xii+496+ 33 plates. (London: The Athlone Press, Universit of London, 1959.) Distributed by Constable and Co. Ltd. 45*s*. net.

UNTIL a relatively short time ago, experienc was the great postgraduate teacher, but th progress of knowledge in the past thirty years ha made it clear that experience alone is no longe enough. Other teachers are necessary, capable o helping medical men and women to implement th new ideas and discoveries in their daily work whatever that may be. Twenty-three of these teacher have prepared their lectures for this book. It is th seventh in a well-known series, and the subject covered are as wide as before. They range fron studies of the growth of viruses to biochemica deficiencies in the whole animal, and include lecture on hæmatology, biochemical genetics, cell regeneration, pharmacology, minutiæ of the eye and ear, th stability of collagen and ageing processes in th female reproductive tract.

The subjects could only have been presented b an equal number of specialists, and they display th complexity of the biological framework as it i gradually becoming revealed to us. No one man ca comprehend more than a fraction of it all in detail but these lectures year by year are mostly simpl enough to allow the interested reader to draw hi own conclusions as to the bearing, if any, they ma have on his daily round. Nevertheless, the spoke word is often easier to follow than the written one and some of these lectures must be studied with car to get the most out of them, and for many peopl time and interest will be the limiting factors.

This series of books is described as a venture i postgraduate education, intended for graduates i medicine looking forward to careers as teachers an consultants in medicine and surgery and their specia branches, as well as for those seeking careers i research and teaching in the medical sciences. This i a very fair appraisal and the sponsors may congratulate themselves on their venture. It is only on of the many now being made in postgraduat education. The short courses lasting from one t three days now being held at the Royal College o Physicians, at Cambridge and in many other parts o Britain, have rather a different appeal, but judgin by their success, whether the proceedings are published afterwards or not, there is evidently a widespread demand for knowledge among specialists an practitioners, and an appreciation of the efforts whic are being made to provide it for them.

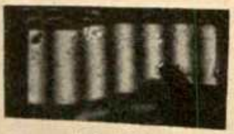
R. A. MCCANCE

INTERFEROMETRIC STUDIES ON SYNTHETIC DIAMONDS

By Prof. S. TOLANSKY, F.R.S., and Dr. I. SUNAGAWA
Royal Holloway College, Egham, Surrey

In a recent article we reported on some findings on synthetic diamonds[1]. Using a very sensitive phase-contrast microscope we observed the following features which do not occur on natural diamonds, namely: (*a*) cubic faces of unusual smooth quality; (*b*) growth spirals on some such cubic faces; (*c*) dendritic diamonds; (*d*) very frequent linear slip lines, especially on octahedral faces (such slip is found in natural diamonds, but much less frequently).

We have now succeeded in obtaining interferogram on quite a large number of synthetic diamond faces. Owing to the micro-character and irregularity of shape of the material at our disposal, we have so far only succeeded with two-beam interferometry, but we are still pursuing attempts to apply multiple-beam methods. We have available both the samples reported upon previously and also a new sample (kindly sent to us by Dr. Bundy of the General Electric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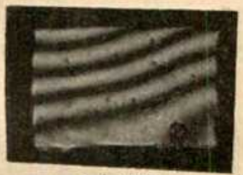
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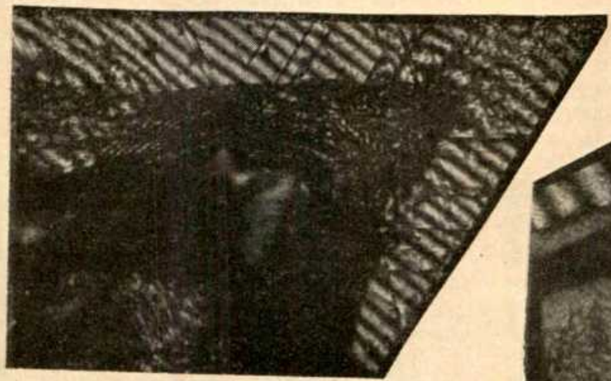
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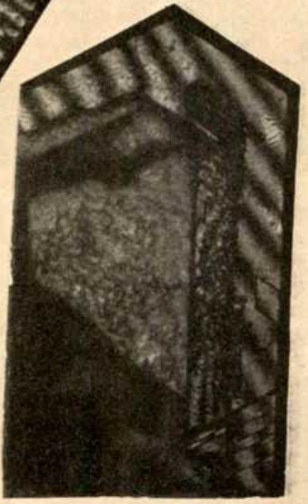
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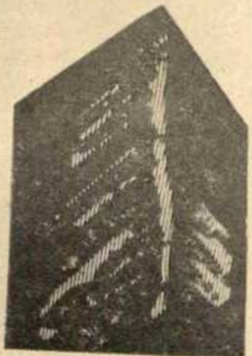
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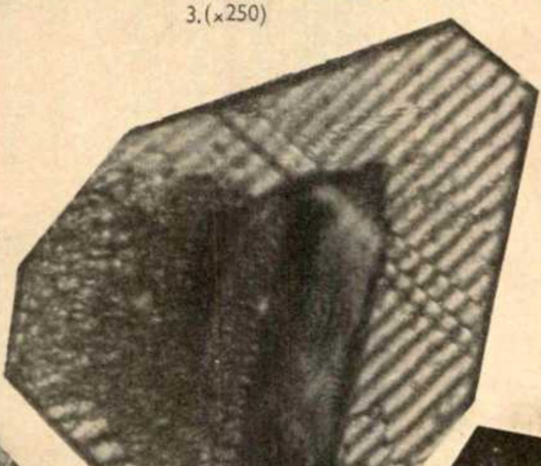
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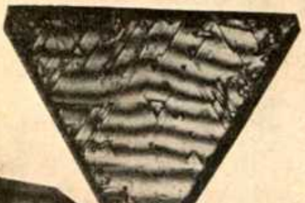
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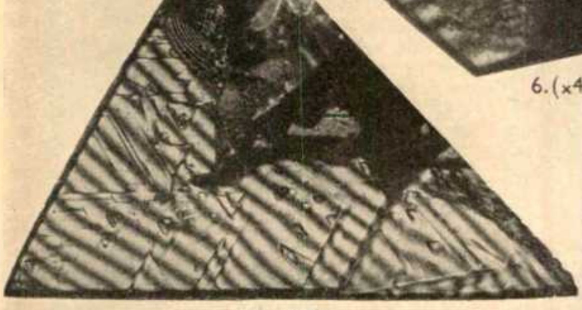
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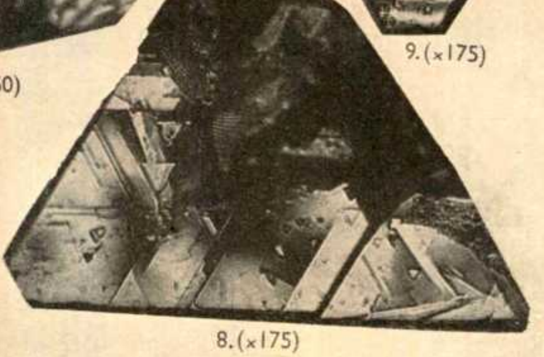
8. (×175)

Co., Schenectady). The older sample reveals many cubic as well as octahedral faces. The crystals in the second sample are perhaps twice as large linearly (some $\frac{1}{5}$ mm.) and are almost exclusively octahedra. Interferograms from the earlier sample will be considered first.

The interferograms were obtained by matching against glass. The fringe visibility found calls for comment. Normally the matching of gem-quality diamond against glass produces fringes of moderate visibility, because of the two very different reflecting coefficients. Here, on the contrary, many of the fringe patterns are of such excellent visibility that it must be concluded that the reflecting coefficients of the diamond surfaces are relatively low, indeed not very different from that of glass. This fringe visibility is particularly noticeable on some, but not all, of the fringe patterns, and it is quite possible that the few cases with poor visibility are due to a nearer approach in reflectivity to the typical natural value. We have no firm evidence as yet as to the possible causes for a reduction in reflectivity, that is, as to whether the defect lies in the crystal or is due to surface film. Figs. 1 and 2 show fringes on two fine smooth cubic faces. The smooth uniformity in the fringe patterns is ample confirmation of our earlier statement that synthetic cubic faces can at times show a remarkable degree of perfection. Fig. 1 shows an excellent plane (within the limitations of the two-beam fringes). The good fringe visibility is worthy of comment. Fig. 2 also shows a smooth, nearly flat, surface, but with a hillock over one corner, about one fringe in height. It is more than likely that this is an unresolved growth spiral, since we have most frequently seen such spirals starting out from corners. Two-beam interferometry cannot be expected to resolve the spirals we have previously described.

We consider next, in Fig. 3, an octahedral face, and this interferogram reveals three commonly found characteristics: (*a*) First is the marked skeletal or hopper character of the face. It reveals a level, fairly plane, parapet or rim and within this is a hollow region. (*b*) Second, the fringes on the rim or parapet show that this region is smooth and of good quality. (*c*) Third, and particularly noticeable, are the crystallographically oriented slip lines, some of which end in the surface.

We have found numerous frequently interesting variations of the hopper or skeletal formation and they are particularly well brought out by the fringe system, in that the upper rim is that which contacts the optical flat. Fig. 4 is one further example from many. There are local variations in fringe visibility here and it is well to remember that the good visibility comes probably from the defective regions, the bad visibility on the contrary being attributable to a more normal reflectivity. Indeed, there is a suspicion on the originals that the regions of good visibility of fringes are patchy and perhaps coated.

A few dendritic crystals have responded to interferometric analysis and part of one such dendrite is shown in Fig. 5. The fringes show that both the 'backbone' and the 'ribs' are flat and smooth, and indeed co-planar, more or less.

We come now to the consideration of the interferograms given by the second batch of crystals, largely octahedra. Here again the hopper character is common, as before, and the slip lines are frequent. Fig. 6, taken with blue light, reveals the hopper character and shows strongly marked slip running right across the crystal, even across the central hollow. It seems fairly certain that the slip here has taken place very late in the history of the crystal. By the employment of different wave-lengths the heights of the steps have been determined. They vary from 500 to 15,000 A.; a good deal more than might have been expected.

Fig. 7 shows a characteristic of the slip lines which we find to be very common, namely the way in which they terminate. Many do so in the surface, and the fringe displacements on either side show quite clearly that the slip step is inclined to the plane of the surface of the crystal. It has, in fact, the character of a screw dislocation.

Finally, we direct attention to the occasional appearance of the familiar growth trigons seen so often on natural octahedral faces. Their visibility, when shallow, is much enhanced by interferometry, in accordance with familiar principles. Thus trigons are clearly seen in Fig. 7, and more so in Fig. 8. Fig. 8 is the same face as Fig. 7; but here we have arranged to have a very high interferometric dispersion by setting the face almost exactly parallel to the optical flat. The result is to create the usual enhanced interferometric contrast and this shows up the trigons well.

The occurrence of well-defined linear trigons on diamonds which have grown very rapidly and which show not the slightest sign of etch or solution effects is in complete harmony with our long-held views that trigons are essentially a growth phenomenon. As so often observed in natural diamonds, so too in synthetic diamonds the trigons frequently 'sit' on a linear dislocation (Fig. 9).

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[1] *Nature*, **184**, 1523 (1959).

BIOLOGY OF THE SOUTHERN COLD TEMPERATE ZONE

IN opening a discussion meeting of the Royal Society, held during December 10–11, which was attended by some twenty invited contributors, including many from overseas, the leader, Prof. C. F. A. Pantin, stressed the importance of understanding natural distribution patterns of animals and plants. The chief problem of to-day is not how to maintain a maximal rate of technological advance, but to predict and control the biological future of the Earth. The study of the development and adaptations of faunas and floras in the past, and of their response to climatic and geological factors, may provide vital information for the future.

The Southern Cold Temperate Zone is the least known of the world's major climatic belts, and information concerning it is widely scattered. Because of the wide separation of its land masses there has been a tendency for the separate consideration of their floras and faunas. This discussion was the first attempt during this century at a general synthesis of the available biological information about the region as a whole.

The Royal Society's interest in the Southern Temperate Zone arose in connexion with the Darwin–Wallace Centenary, which it celebrated by sending a five-man expedition to southern Chile, in the southern summer of 1958–59. This party carried out geological and ecological surveys, and made observations on species with a disjunct distribution linking Chile and New Zealand (M. W. Holdgate*). Originally the Royal Society had been interested simply in this trans-Pacific relationship; however, as a result of the expedition, it was realized that this should be considered within the broader context of the whole southern circumpolar region.

The Southern Cold Temperate Zone includes a large proportion of ocean: the climate of its land masses is therefore unusually closely related to marine temperatures and currents (G. E. R. Deacon). Southwards the Antarctic Convergence provides a sharp and fairly constant boundary: the land within it, like Antarctica itself, South Georgia and Bouvet Island, is largely ice-capped. Northwards, the Sub-Tropical Convergence fluctuates more widely but includes nearly all the temperate southern islands. Southern South America, New Zealand and Tasmania are the only truly continental southern temperate areas.

According to C. J. F. Skottsberg, many plant families must be regarded as Palæo–Antarctic, and are still largely confined to the southern cold temperate region (for example, Eucryphiaceae, Gunneraceae, Escaloniaceae, Proteaceae). Some members of these are strictly continental in distribution, but even those genera reaching oceanic islands fall roughly into either 'balanced', 'South American', 'Neozelandic–Australian' or 'Afro–Indian' groups. *Acaena*, widely spread around the zone, exemplifies the first type, *Pernettya* and *Fuchsia* the second, and *Hebe*, *Coprosma* and *Nothofagus* the third. Viewing the pattern as a whole, the paths of migration of these groups seem to radiate from the Antarctic. Their present distribution does not seem readily explicable in terms of trans-oceanic dispersal, and seems to accord best with an initial segregation in Antarctica itself and subsequent outward spread (C. J. F. Skottsberg and G. E. DuRietz). Such a hypothesis is strongly supported by geological evidence (R. A. Couper). In the late Cretaceous and early Tertiary *Nothofagus* species and Podocarpaceae were abundant in west Antarctica (Seymour Island), Australia, New Zealand, Kerguelen and southern South America. Many species disappeared from the southern areas in the late Pliocene, at which time a northward shift seems to have occurred. These plants have never been found as fossils in the northern hemisphere or the tropics, and their present extension into New Guinea and Indonesia is regarded as recent.

Vegetationally, southern Chile and New Zealand, as the two main southern temperate areas, have much in common (E. J. Godley). In both the wet, oceanic, western regions are dominated by evergreen rain forests in which *Nothofagus* species are prominent. The New Zealand forests are, however, much richer in species. Other differences include the lack of deciduous beeches in New Zealand (there are seven species in Chile), and the greater importance of podocarps as forest dominants there. Only the open 'Magellanic moorland' of south-western Chile is regarded as truly Sub-Antarctic. Bog vegetation of the 'carpet' type, formed by species of *Astelia*, *Donatia*, *Gaimardia*, *Oreobolus*, and similar plants, rather than *Sphagna*, prevails in this region and also in the New Zealand Sub-Antarctic islands, and is considered an important indicator of Sub-Antarctic conditions.

The southern islands also show fairly clear similarities in the structure of their vegetation. Only the northernmost (Tristan, Gough, Amsterdam, Juan Fernandez, Auckland, Campbell) have scrub, which is always of microphyllous prostrate evergreen trees bearing a heavy lichen and bryophyte epiphytic cover and with an under-storey rich in ferns (N. M. Wace). Large tussock-grasses (usually *Poa* spp.) form a characteristic formation on steep sub-maritime slopes throughout the zone. 'Wet heath' formations of Gramineae, Cyperaceae, ericoid shrubs and bryophytes form a higher belt, with areas of wind-eroded feldmark and montane grassland highest of all. These Sub-Antarctic lowland formations have a striking physiognomic resemblance to the vegetation of high tropical mountains (N. M. Wace and C. Troll); a fact which may be related to the small annual temperature variation in both regions (C. Troll).

In the Quaternary, southern Patagonia had at least three major glaciations. Pollen analysis reveals a post-glacial pattern of climatic and floristic change strikingly like that in the northern hemisphere (V. Auer). There is evidence that such processes are still going on, and the forest-steppe boundary in Tierra del Fuego seems to be fluctuating rapidly. Volcanic ash bands occur at constant horizons throughout most of Fuego-Patagonia, the three latest being dated at 9,000, 5,000 and 2,200 years before the present. These may perhaps be correlated with climatic changes (H. H. Lamb), and fluctuations in the position of the west wind belt and ocean currents are certainly to be expected to follow changes in temperature. A striking agreement in dating exists between the third Patagonian eruption and an ash band in the peats of Gough Island in the mid-Atlantic (U. Hafsten). However, pollen analyses of the Tristan–Gough group peats shows that the present mild oceanic climate, and the present vegetation, has persisted there for at least the past 5,000 years. The main vegetational changes seem to have followed human discovery (U. Hafsten).

The terrestrial fauna of the southern Chilean forests becomes progressively impoverished as one passes from the rich, warm-temperate, Valdivian type into the poorer Magellanic forests of the south, but the latter contain few species absent from the former (G. Kuschel). The western Magellanic moorlands are even poorer faunistically; on the other hand, the open, semi-arid steppes of eastern Patagonia are comparatively rich. Many southern Chilean invertebrate groups have a disjunct distribution in South America and Australasia, paralleling that of the plants, and some of the insects belong to orders which are strictly continental and known from no oceanic island (for example, Plecoptera, Megaloptera) (G. Kuschel and M. W. Holdgate). An outward migration from the Antarctic in the Mid-Tertiary may provide the best explanation of their present range. The terrestrial fauna of the southern oceanic islands, even of relatively warm examples like the Tristan–Gough group, is even more strikingly impoverished. Only 132 terrestrial invertebrates have so far been recorded from these islands. Many of these show a strong South American affinity, and the insects are believed to have been derived predominantly from that continent by airborne immigration in the prevailing westerly winds (M. W. Holdgate). Occasionally South American Lepidoptera and land birds appear as stragglers in the islands. The Tristan group also shows slight faunistic affinity with St. Paul and Amsterdam, but none with the colder and more southerly oceanic islands. These do, however, share something of a truly circumpolar fauna with relations to South America and New Zealand. Most of these islands are believed to be young, and their faunas likewise. In the Tristan–Gough group this hypothesis is supported by botanical evidence (N. M. Wace).

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* The names appearing in brackets in the text of this article are those of the contributors to the meeting whose papers are here summarized. It is hoped to publish all the major Contributions in the *Proceedings of the Royal Society* (B).

Within the littoral marine fauna, it is possible to recognize a distinct southern circumpolar element paralleled in the algal flora (G. A. Knox). Many of the species are dominants and give a characteristic aspect to southern temperate shores. A schematic classification of these zonations can be derived, although distinct species groups are peculiar to each southern continent. Africa, despite an upwelling of cold Sub-Antarctic water off the south-west coast, has little truly southern cold-temperate faunistic relationship (T. A. Stephenson), a fact paralleled in its terrestrial invertebrate fauna (P. Brinck) and its flora (R. S. Adamson). In planktonic organisms, also, the southern waters have circumpolar, clearly zoned forms which show abrupt specific limitations near the convergences (N. A. Mackintosh). The Antarctic penguins also show somewhat of a circumpolar distribution with a radiation to distinct and geographically limited species farther north. In contrast, the shelf fauna of the Antarctic itself has a very high (70–80 per cent) incidence of endemism, and while many of its component species spread all around the continent they show no clear relationship with the animals of the other southern land masses (N. A. Mackintosh).

Oceanic birds are of profound ecological significance in the south, since they breed in vast numbers on many islands. One major vegetation type—tussock grassland—may be in part a biotically induced formation (N. M. Wace). Yet despite their abundance, these birds have so far been imperfectly studied and the validity of their taxonomic definition is still questionable in many cases (R. C. Murphy). Many species make great migrations, and may act as dispersal agents for seeds and invertebrates: this role may be filled especially by juvenile petrels, which often fly while still bearing down contaminated from the nest burrow (R. A. Falla).

There is no doubt that the Southern Cold Temperate Zone possesses many features as a whole. Despite wide separation, there are many floristic and faunistic similarities between southern America and New Zealand and Australasia, and this applies also to the structural features of almost all the southern cold–temperate vegetation types. There is geological evidence which accords with that of present distribution in suggesting a common origin for such groups in a temperate Antarctic during the Tertiary. In contrast, no terrestrial vertebrates have a distribution of the true southern cold–temperate type (P. J. Darlington), and indeed, except for birds, land vertebrates seem to be largely absent from the region. Those that do occur appear to be recent immigrants from the north. Ecological rather than distributional factors may underlie this anomaly. It is evident that many features of this zone reflect the exacting requirements of the high-oceanic climate. The species-poverty of the southern islands and of south-west Chile no doubt depends in part on dispersal difficulties and upon recent ice-retreat, but it may well also derive from the narrow and specialized range of habitat conditions. The Southern Cold Temperate Zone indeed emerges as an area with a highly characteristic biology, and with a climate more profoundly influenced by the temperature and circulation of the sea than any other major world zone (C. F. A. Pantin). It is apparent, however, that much more work remains to be done there. Native faunas and floras are fast vanishing before the onset of northern hemisphere species introduced by man; yet there is virtually no southern island or mainland area which can be said to have received adequate study in all respects.

M. W. Holdgate

AN INTERNATIONAL TAGGING TEST WITH SALMON (*SALMO SALAR* L.) SMOLTS

By A. SWAIN and P. E. CARADINE

Freshwater Fisheries Laboratory, Ministry of Agriculture, Fisheries and Food

FOR many years various experiments have been carried out in Great Britain and other European countries to investigate the migratory habits of the Atlantic salmon (*Salmo salar* L.). Much of the work has been undertaken to obtain further information about the movements of salmon in the sea, the proportion of smolts returning as adult fish and the percentage of spent fish surviving to spawn in following years, and in recent years thousands of salmon smolts have been tagged in Britain and Scandinavia.

Nearly all these experiments have involved the marking or tagging of the fish in some way. Numerous methods of tagging have been tried both with internal tags and external ones attached to various parts of the fish's body. The tagging of salmon smolts presents special difficulties because of the difference in the size of the fish between tagging and recapture. The lengths of most of the smolts tagged lie within the range 12–18 cm.; but when the fish are recaptured after spending one, two or more years at sea, the lengths will be between 50 and 100 cm.

or even longer. It follows that a tag which is attached to such a small fish must not restrict its freedom of movement unduly, but at the same time the tag must be such as to remain on the fish for a number of years. Also, in most instances, the tag must be clearly visible when the adult fish is caught. Although the type of tag has varied considerably, most of the workers have attached it near the base of the main dorsal fin of the fish.

In Great Britain, the number of tagged smolts being recaptured as adult fish has rarely exceeded 2 per cent. Norwegian returns have been somewhat lower, whereas in Sweden, the average figure has been between 5 and 10 per cent, with even higher figures for some of the larger smolts tagged. These high returns have been obtained both with Baltic salmon, where the fish are caught commercially on the feeding grounds, and with west-coast salmon, where they are caught along the coast near the mouths of the rivers.

The efficiency of different tagging methods was discussed at a symposium held by the Salmon and Trout Committee at the meeting of the Conseil International pour l'Exploration de la Mer, in October 1958. It was proposed that there should be an interchange of tagging methods. As a result, during April and May 1959, a number of Scottish smolts were tagged by Scottish, English and Swedish workers, using their own tags and techniques. Later on in May we visited Norway and Sweden in order to apply our English methods to the tagging of Scandinavian smolts.

All the smolts tagged in Norway and Sweden during this experiment were hatchery-reared and were two years old. We used two types of tag, one of which was a small silver plate, the other a small square of 'Ivorine' (celluloid)[1]. These tags were serially numbered, and for both types a short length of fine silver wire was used as a means of attachment. The Swedish tag (Carlin tag)[1] consisted of a small piece of cardboard coated with celluloid, with a message printed on one side of the cardboard. A serial number appeared on the other side. The tag is attached by means of a moveable hinge to a short length of soft stainless-steel wire which in turn is hinged to another stainless-steel wire, both ends of which are separately inserted into the body of the fish and twisted together on the other side. The tag used by the Norwegians is called the Lea hydrostatic tag[1] and is used in Britain for tagging adult fish. It consists of a small sealed plastic cylinder containing a printed message. It was attached to the fish by means of a bridle of soft stainless-steel wire. In all cases, hypodermic needles were used to facilitate the passage of the wires through the fish. A single needle was employed in the English and Norwegian techniques, a double needle in the Swedish method. The English and Norwegian workers attached the tag to the base of the anterior end of the main dorsal fin, so that the tag is carried on a loop above the back of the fish. The Swedish workers attached the tag to the body of the fish immediately below the dorsal fin. Thus, with the English and Norwegian methods, a small loop of wire extends over the back of the fish, whereas with the Swedish method there is no such loop. The Norwegian and Swedish workers anæsthetize the smolts with a 2 per cent solution of urethane for the tagging operation. The English workers do not normally use anæsthetics, but the fish are tagged in running water using a special holding block.

In Norway, more than two thousand smolts were tagged by the three different methods at the Lundamoen Hatchery near Trondheim. The fish were all released into the Gaula River. In Sweden, the production of electricity is based almost exclusively on the utilization of water-power, and a series of dams is being built on the main salmon rivers flowing into the Baltic Sea. The effect of these installations would be to destroy large areas of natural spawning grounds for salmon. The seriousness of such a situation would appear even more acute when it is realized that the particular value of the Baltic salmon fishery lies in its complementary relationship to the very important herring fishery in these waters. It has been decided, therefore, to substitute for the natural spawning grounds a system of hatcheries and rearing stations whereby the run of salmon can be maintained artificially by the annual release of a sufficient number of hatchery-reared smolts. In order to discover whether this system could be worked economically the Swedish authorities have tagged large numbers of artificially reared smolts during recent years.

A comparison of Swedish and English tagging methods was carried out at three stations on the east coast. The first batch of smolts was tagged at Galtstrom just south of Sundsvall. Fish reared at this particular station have normally produced a very high percentage of recaptures. The smolts were larger than normal, averaging 16–18 cm. in length and in excellent condition. More than four thousand smolts were tagged at Galstrom and all the fish were released into the nearby River Ljungan.

Another collection of smolts was tagged at Bergeforsen, near the mouth of the Indalsalven, one of the large rivers flowing into the Gulf of Bothnia north of Sundsvall. Here is situated one of the largest Atlantic salmon hatchery and rearing stations in the world. By means of a device consisting of batteries of plastic trays, more than two and a half million salmon eggs can be brought to the 'eyed' stage in quite a small room. The rearing station is constructed for the production of about four hundred thousand smolts of salmon and sea trout reared for two or three years. About 1,200 smolts were tagged at Bergeforsen and the fish were released into an artificial channel from which they could migrate into the main river at any time.

The third batch of smolts were tagged at Lulavan, situated on the Lule älv near the port of Luleå in Northern Sweden. The installation at Lulavan was merely a tagging station; but the history of the rearing of these smolts was most interesting. Although the salmon ova were from the north, the fish had been reared at Munka-Ljungby on the west coast of southern Sweden, where growth was more rapid. At the smolt stage, the fish had been transported back to the north by road tanker, a distance of 1,600 km. More than six hundred smolts were tagged at Lulavan and the fish were released into the main river.

In the spring of 1960, it is expected that Scottish, Swedish and Norwegian workers will tag smolts in an English river. Here, the fish will not be hatchery-reared but will be tagged during their natural migration out to sea.

It is hoped that some tagged fish will return in 1960; but it is clear that a statistical comparison of the methods of tagging employed in 1959 will not be possible until the end of the 1961 season, for only then will the major part of the 1959 smolt crop have been harvested.

[1] *J. Cons. Intern. Explor. Mer*, 9, No. 2 (1953).

RESEARCH METHODOLOGY AT THE LONDON SCHOOL OF ECONOMICS

By Prof. M. G. KENDALL

Director of the Research Techniques Division

RESEARCH techniques in the social sciences grew up in a very haphazard fashion; and some of them are so rudimentary that they can scarcely be said to have developed at all. After the Second World War it was plain that, if effort was not to be misdirected and wasted, some systematic assault on problems of methodology in the social sciences was necessary. With the aid of a grant from the Nuffield Foundation the Research Techniques Division at the London School of Economics and Political Science was set up to study such problems. It has no precise terms of reference and ranges over a wide field; but, broadly speaking, it is concerned with the gathering and analysis of information and the testing of scientific hypotheses in economics, sociology, social psychology and related fields.

The Division began with a grant of £2,000 a year, a part-time director, a secretary and two research assistants. At the present time, having just celebrated its tenth birthday, it has a budget of about £30,000 a year and a staff, full-time or part-time, of more than thirty. These figures will probably increase by 50 per cent over the next two or three years. Most of the funds come from outside sources. Some of the staff are full-time research officers, others are teaching members of the School staff who are associated with particular projects and others again are outside consultants. The Division has its own computing unit and library.

Methodology of Social Surveys

The elicitation of social and economic facts is itself a complicated subject. It may involve the setting up of new systems of scale or measurement unknown to the physical sciences. Even the 'mere' collection of data involves extensive methodological study, especially in the field of social surveys. From the planning of a survey, through the design of the questionnaire, the selection of the sample, the handling of the interview situation, the coding of responses and the final analysis, there lies a long chain of procedure which can introduce error at almost any point along its length. The Division carries out studies of the nature and extent of such errors in order to devise methods of reducing them and of validating the final results. It has, for example, published studies of the sampling problems involved, of the effect of using amateur interviewers, of coding errors and of the way in which response varies with the type of question asked.

More recent studies embarked upon cover the adequacy of mail questionnaires, memory bias in respondents and the characteristics of persons who volunteer for panels. On the sampling side, although the drawing of samples of human beings in the United Kingdom is now fairly well understood, there remains a number of intractable problems for study: the sampling of rare events, the sampling of firms or retail shops, and, overseas, the sampling of migrant populations or under-developed areas.

Dynamic Economics

Many natural scientists who have taken their interest in economics so far as to read a text-book must have wondered whether it is not possible to reduce the subject to the much more formal lines with which they are familiar in their own discipline. It is not, I think, true that economics can be 'turned into a science' in the sense that the methods of the natural sciences can be taken over unchanged and applied in a new field. But I believe it to be true (and this belief forms the starting-point of the Division's work on economics) that with suitable new techniques or adaptations of old ones the behaviour of economic systems is capable of far more exact study than has been attempted in the past. Under the inspiration of Prof. A. W. Phillips, and with the help of a substantial grant from the Ford Foundation, the Division is engaged in a large-scale attack on this problem.

The first requirement is to bring the postulated relationships of economics to an explicit form for quantitative study—to write down the equations. This is itself far from easy. The second problem is to write down the model of the system and to investigate its behaviour. The third stage is to fit such models to real life, which involves intricate problems of estimation and testing of hypotheses. The final stage is then to use such models in the regulation of economic systems.

It is particularly interesting that the main stimulation for such work has been furnished by electrical engineers, who have realized the formal similarity between economic systems and the feed-back networks of control engineering. The economic problem, of course, is more complicated, especially in regard to the amount of random variation and unknown time-lags to which economic systems are subject. The Division has been working in this field for several years, and has now reached the point where most of the economic relationships can be formulated, the estimation problems solved and the numerical computations carried out on an electric computer. The properties of artificial models are now being studied in preparation for the next phase, which is the study of actual portions of the economy.

Operational Research

Operational research—the science of making the best of limited resources in complex man–machine situations—is a comparatively new subject in both emergence and scope. With the aid of a small grant from the Department of Scientific and Industrial Research, the Division has spent two years in self-education, preparing bibliographies and running a fortnightly seminar attended by operational research workers from outside the School. It is now proceeding with research studies on some of the methodological problems involved in such topics as linear programming and queueing theory, and members of the Division are engaged in actual operational research for certain firms. One collateral development has been the setting up of systematic courses at the London School of Economics for postgraduates drawn from industry. It is by now quite plain that this side of the Division's work will expand substantially in the years to come.

Research on Statistical Method

A great many problems coming to the Division's attention are necessarily of a statistical character, and attempts are made to solve such theoretical statistical problems as arise. The work covers a very large field. At the mathematical end, for example, it has included the tabulation of the distribution of the characteristic roots of a random-dispersion matrix. A considerable number of studies have been published on distribution theory, estimation and inference, the theory of statistical relationship, time-series and stochastic processes. Such work, though in the main theoretical in character, usually stems from problems of analysing complicated and imperfect data such as arise in multivariate systems.

Bibliographical Work

A great deal of work within the Division's sphere of interest, as in scientific fields generally, is concerned with ascertaining what is being done elsewhere. A number of individual bibliographies have been published, for example, on queueing theory and on the life testing of industrial equipment. Some special studies have also been undertaken. For example, a "Dictionary of Statistical Terms" has been prepared on behalf of the International Institute of Statistics and Unesco; a new *Journal of Statistical Abstracts* has been founded; and the Division is on the point of going to press with the first volume of a comprehensive bibliography of papers on statistical method and probability from the sixteenth century up to the end of 1958 which will ultimately comprise about 25,000 titles.

Team Work

It is a characteristic feature of much of the Division's work that its problems require collaboration between workers in several different fields. Problems of survey research, for example, need mathematicians and statisticians at the sampling stage; psychologists at the interview stage; numerical analysts and statisticians at the summarizing stage; and sociologists or economists at the interpretative stage. Dynamic economics requires—in addition to economists—electrical engineers, numerical analysts and mathematicians. The members of the Division are, in consequence, drawn from all sorts of disciplines, and it frequently calls on the wide resources of the teaching staff at the London School of Economics as well as on those who are formally attached to it. The consequent interactions have done much to contribute to the Division's development and, I hope I may say, to its successes.

OBITUARIES

Sir Henry Tizard, G.C.B., A.F.C., F.R.S.

It is often said that you cannot keep a good man down, but you only have to look at the early history of many eminent people to realize how much they owe to chance. Tizard was thirty-five before he took the step which enabled him to develop his powers to the full. I believe that it was then that he took the quite conscious decision to become a V.I.P. He certainly achieved his greatness by his own efforts.

Tizard's forebears were all connected with the sea in some way or another and he, himself, was destined for the Navy, but failed because of eyesight. His mathematics gained him an exhibition at Westminster School, and at the outbreak of the First World War he was a Fellow of Oriel College, Oxford, having got there by way of a first-class degree in natural sciences at Magdalen, a year at Berlin under Nernst and two years at the Davy Faraday Laboratory of the Royal Institution.

When the War actually broke out, he was in Australia attending the meeting of the British Association. He returned as quickly as he could and joined the Royal Garrison Artillery, where he got himself into some difficulties owing to his highly unorthodox methods of training recruits. This, in fact, was not really his 'cup of tea' at all and he quickly succeeded in getting transferred to the Royal Flying Corps By 1917 he was captain-in-charge of all experiments on aeroplanes at Martlesham Heath and was afterwards appointed as deputy to Bertram Hopkinson at the headquarters of the Ministry of Munitions. He carried on after Hopkinson had been killed in August 1918 until he was demobilized in the spring of 1919, when he returned to Oxford.

His experiences during the War had led him to realize the immense importance of the application of science to military affairs. He decided that he would never be outstanding as a pure scientist, and Oxford, therefore, did not give him the scope that he needed to develop his talents. Towards the end of 1920 he joined the Department of Scientific and Industrial Research, and in 1926, at the age of forty-one, became secretary of the Department in succession to Sir Frank Heath. In the same year, he was elected a Fellow of the Royal Society.

In 1929 he was appointed rector of the Imperial College of Science and Technology—a post which he held for thirteen years. He did, in fact, a very great deal for the College, but, during this period, he did a great deal more in other directions.

Ever since the First World War, Tizard had maintained his interest in aeronautics. He had been chairman of the Royal Aeronautical Society in 1924, and in 1933 he was appointed chairman of the Aeronautical Research Committee—a post that he held for ten vitally important years. However, the work for which I shall always give Tizard the highest marks in that period was the job that he did as chairman of the Committee for the Scientific Survey of Air Defence which started work at the beginning of 1935. Tizard's colleagues on this body were originally A. V. Hill, P. M. S. Blackett and H. E. Wimperis, with A. P. Rowe as secretary. I shall always believe that this body, and particularly Tizard himself, were the people who made it possible for Watson-Watt to develop radar, which was the biggest contribution to the winning of the Second World War. You cannot read the accounts of the development of radar written by such varying authorities as A. P. Rowe, Lord Swinton and Watson-Watt himself without coming to the conclusion that it was Tizard's drive from the outside which ensured that radar was developed at such rapid speed—and not only developed, but also deployed in a usable form.

As the War progressed, Tizard served in the Ministry of Aircraft Production and also as the first—and last—external civilian member of the Air Council. In August 1940 he led a British mission to the United States with the object of disclosing all our scientific secrets. He took with him in his pocket a sample of the magnetron—the thermionic device, perfected in Britain by Randall and Boot, which really made radar work. One American historian said of this visit: "When the members of the Tizard mission brought the magnetron to America in 1940, they carried the most valuable cargo ever brought to our shores. It started the whole development of micro-wave radar and constituted the most important item in reverse lease-lend". In the years after the War, when information on American military research was hard to obtain, Tizard was not slow to recall his war-time mission.

In 1942 Tizard resigned from the Ministry of Aircraft Production and the Air Council to become president of Magdalen College, Oxford; but, although he now had no formal Government appointment, he still continued to advise informally on various scientific problems, and in 1943 he went out to Australia to advise the Australian Government. It was during this trip that he bearded General MacArthur in his den and shattered the morale of the General's aides by flatly refusing to regard him as anything but an equal. Incidentally, he also won a case of sherry off the General by a bet on the length of the War.

By 1946 Tizard was sixty-one. He was very comfortably ensconced as president of Magdalen and it looked as though the excitements were over. His greatest work was, in fact, still to come.

One of the major lessons of the Second World War was the vital necessity to apply science to all forms of Government activity, and this could only really be done provided the right type of scientist was brought into full collaboration in both the formation and implementation of policy. The Services learned this lesson very thoroughly indeed during the War and, as a result, the old Deputy Chiefs of Staff Committee was replaced by a new body in the Ministry of Defence known as the Defence Research Policy Committee. The Committee consisted of those responsible in the Service Departments and the Ministry of Supply, both from the operational and from the scientific angle, for the formulation of requirements for new weapons and for research and development. Its chairman was to be a scientist of high standing appointed for a period of years. At the same time, on the Civil side of Government, a committee known as the Advisory Council on Scientific Policy was also set up. Tizard was persuaded to assume the chairmanship of both these bodies; and, although from 1946 he served full-time in the Ministry of Defence, he gave a great deal of attention to the work of the Civil Council.

Between 1946 and 1950 Tizard laid the real foundations of co-operation at the top level between the scientist and the military authorities. His own position and effectiveness did much for the recognition in other fields of the need for, and value of, scientific advice. I do not believe there was any other person who could have done this with anything like comparable success. For one thing, in spite of the fact that neither he nor anybody else regarded him as an outstanding pure scientist, his standing in the scientific world was extremely high. Even by 1946, five universities had awarded him honorary degrees, and in subsequent years five more followed suit. For another, he had shown, in the days before the Second World War, his capacity to see what was important, from the national point of view, to develop and, perhaps even more, the ability to get other people in high places to accept his view. He inspired—and in this he was almost unique among the leading scientists of the day—immense respect and confidence among officers of all the Fighting Services. He had a very genuine appreciation of, and sympathy with, their problems and they all knew he wanted to help.

He dominated any committee of which he was chairman, not—as so many other people try to do—by talking all the time, but by his skilful appreciation of the characteristics of the people sitting with him and of the appropriate moment at which to intervene. Sometimes, these interventions took the form of an exhibition of bad temper. Most people thought this was spontaneous; but, in point of fact, it was most carefully rehearsed, as those who had to brief him for the meeting well knew. In those days, his moments of real anger were quite rare, and they almost invariably arose out of his unceasing warfare with what he regarded as administrative stupidity.

He was, perhaps, at his most impressive when presiding over the periodic meetings of the Commonwealth Advisory Committee on Defence Science, to which scientists and serving officers came from all over the Commonwealth. Here, he inspired a slightly awed but affectionate respect. He was, of course, a brilliant conversationalist, and the entertainments which accompanied these Commonwealth meetings gave him opportunities which he seized to the full.

He was on excellent terms with the Chiefs of Staff, particularly in the earlier days. As time went on, however, he began to be regarded as the 'Great Man', and his visits to the Chiefs' meetings became rather more formal occasions. There was, in fact, something rather magisterial about his manner, especially on any formal occasion. Even on an informal occasion, he was not a man with whom one took undue liberties. Nevertheless, to those who worked with him closely, he was a very real friend and by no means difficult.

In 1950 his health began to fail and by doctor's orders he had to slacken off a great deal. He did not take kindly to this change, but there was no doubt that he began to get tired rather quickly and then became a little bad-tempered. He recognized this, however, and I can only remember one occasion which had unfortunate results. By 1952, Tizard had begun to lose some of his confidence in himself. He thought that his health had deteriorated to such an extent that his useful working life was over, and he therefore resigned from the public service. There is no doubt that a measure of the old zest and vitality had gone out of him.

He was not, however, a man who could be idle, and for the remainder of his life he occupied himself as a director of Glaxo Laboratories, Ltd., and other public companies. In spite of many attractive offers, he refused to join any commercial organization primarily concerned with the Defence effort. He took the view—and I am sure rightly—that he had been in far too central a position in the defence field to allow him to make his special knowledge available to a single company engaged in such work.

His health improved appreciably after he retired, and right up to the day of his death he was the most delightful and inspiriting dinner companion. A typical form of activity into which he entered with all his usual enthusiasm was the year he spent in 1955–56 as Prime Warden of the Goldsmiths' Company. I am sure the Company benefited from his wisdom.

No account of Tizard would be complete without some reference to his quarrel with Cherwell. This originated in 1936 over the Committee on the Scientific Survey of Air Defence, and eventually got so bad that the two men could not endure in silence each other's presence in the same city. This, indeed, was the only matter on which I ever found Tizard really unreasonable, but Cherwell was worse. In fact, in this matter, both these eminent gentlemen behaved like a couple of spoiled children. The tragedy of it was that this quarrel prevented Tizard gaining the confidence of Winston Churchill when the latter returned to power in 1950.

By any standards, Tizard was a great man who made a large contribution to the nation's affairs. He had a very real foresight and was rarely proved wrong. I am still waiting for someone to take seriously the warning which he issued towards the close of his presidential address to the British Association in 1948. He knew little about agriculture in detail, but, as with all the other subjects he touched, he had a wide view of the necessities. "I shall predict", he said, "that unless the prevention of disease among plants and animals and all other scientific problems of the supply of food are studied on the same kind of scale by men of similar calibre as are the problems of human health, chaos and misery will result". How right he was.

F. Brundrett

Dr. Mary D. Waller

Dr. Mary Désirée Waller, who died at the age of seventy-three on December 11, was widely known for her precise and beautiful work on vibrating plates and as an outstanding teacher of physics. She came of distinguished scientific ancestry, her father being Augustus Désiré Waller, who first showed that the electric currents set up by the beating of the human heart could be recorded, and her grandfather Augustus Volney Waller, whose name is commemorated in 'Wallerian degeneration'.

Educated at Cheltenham and Bedford College for Women, as a young woman she was appointed demonstrator in the Physics Department of the London (Royal Free Hospital) School of Medicine for Women, a Department of which she later became head and in which she continued her research after her retirement in 1947. It was in 1932 that she first came into prominence by her discovery that metal plates could be thrown into regular vibration by the suitable application of a block of high-density carbon dioxide. It shows great scientific percipience that her discovery derived from the fact that an itinerant vendor of ice-cream had directed her attention to the way in which a bicycle bell rattled when in contact with ordinary solid carbon dioxide.

With this solid carbon dioxide technique, the mode of action of which she explained, she carried out extensive and precise studies of the modes of vibration of metal plates of square, rectangular, circular and elliptical form, illustrated by beautiful photographs, the series recording the vibrations of a square plate being particularly striking. Her last published work included some interesting observations on the ridges formed by particles of different size and shape in the sound field between a vibrating plate and a parallel horizontal surface. At the time of her death she had nearly completed a book dealing with her work on vibrating plates, the appearance of which has been announced by the publisher. It is to be hoped that a colleague will finish the work and see it through the press.

Dr. Waller was a woman of great integrity and kindliness, whose brightness and charm made her a welcome figure in scientific, and other, circles. As an accomplished teacher she endeared herself to generations of students in her Department. She was devoted to the subject that she made her own, but her interests were wide and she was a delightful conversationalist. She lived for many years with her friend, Dr. Bertha Turner, at a charming house in the neighbourhood of Regent's Park, in which she died.

E. N. da C. Andrade

Mr. Cyril L. Collenette

Cyril Collenette, who died suddenly on November 2 at the age of seventy-one, devoted most of his life to the study and service of natural history. His boyhood days were spent in the stimulating surroundings at Woodford, then a country parish in the heart of Epping Forest, and this early bent for the pursuit of Nature's secrets and offerings was never to leave him. He became a world-wide traveller naturalist in such places as Malaya, French Guinea, the Matto Grasso and British Somaliland, making valuable additions to collections, especially of insects.

Collenette's main life's work began and ended at the British Museum (Natural History), South Kensington, as an associate and world authority on the large family of moths, the Lymantridae. Apart from his museum work, he was a keen spare-time botanist and bird observer, ever willing to pass on his experiences and recordings to such bodies as the British Trust for Ornithology (he was one of its earliest ringers), and the London Natural History Society (a member since 1907 and one-time president). He was on the Committee for the Birds of the Royal Parks, as well as on that of the London Natural History Society during the compilation of its book, "Birds of the London Area".

His own books include "A History of Richmond Park", written during 1937 when living at Richmond, and "Sea-girt Jungles", written on his return from a long Pacific sea voyage on the *St. George*, a sailing vessel which visited such places as the Marquesas, Easter Island and the Galapagos. He told me that this was one of the most exciting and rewarding episodes of his natural history career.

His colleagues at South Kensington will miss their tall and friendly companion, and all who sought out his advice and friendship will mourn an ever-willing helpmate and kindly friend. Alfred Leutscher

NEWS and VIEWS

Vice-chancellor of the University of Monash: Prof. J. A. L. Matheson

PROF. J. A. L. MATHESON sailed for Australia before Christmas to take up his appointment as vice-chancellor of the University of Monash, Melbourne, and to begin his exciting and responsible task of building a new University. Matheson, who was educated at Bootham School, York, succeeded the late A. H. Gibson as Beyer professor of engineering in the University of Manchester in 1951, having been lecturer in civil engineering in the University of Birmingham from 1938 to 1946 and professor of civil engineering in the University of Melbourne from 1947 to 1950. During his tenure of the Beyer chair at Manchester, Matheson re-organized the engineering courses and planned new laboratories to house the post-war expansion. In addition, he played an important part in the wider field of university building and planning. He directed research into various aspects of theory of structures, including the use of scale-models and the load-carrying capacity of rigidly jointed triangulated steel frames. It was largely due to his guidance that so much has been done in Manchester to further the use of electronic digital computers for the analysis of complex structural problems. His work culminated earlier in the year in the publication of his book, "Hyperstatic Structures", which is impressive for its scope and scholarship.

Civil Engineering at Manchester: Prof. M. R. Horne

DR. M. R. HORNE, lecturer in the Department of Engineering, University of Cambridge, has been appointed professor of civil engineering in the University of Manchester and will take up the post next summer. Dr. Horne was born on December 29, 1921, and was educated at Boston Grammar School and Leeds Grammar School before going to St. John's College, Cambridge, in 1939. He had a distinguished academic career, obtaining honours and the Archibald Denny Prize in the Mechanical Sciences Tripos, 1941. He was awarded the John Winbolt Prize for an essay on an engineering subject in 1944. During 1941–45 Horne worked for the River Great Ouse Catchment Board as an assistant to the District Engineer, Bedford, surveying and carrying out reconstruction of weirs and sluices. He returned to the Department of Engineering, Cambridge, in 1945, joining the team which was then being assembled to continue the work, started before the War in Bristol, on the plastic behaviour of steel structures. He obtained the Ph.D. degree in 1950. In 1951 he was appointed an assistant director of research on the introduction of the postgraduate course of instruction on structures and materials, the first such course to be introduced by the Cambridge Engineering Department. Horne transferred to the post of University lecturer in 1957, in which year he was elected a Fellow of St. John's College. In the fifteen years that Horne has been back in Cambridge he has played a prominent part in the development of the theory of the plastic behaviour of structures and of the design method based on it.

Director of the Forest Products Research Laboratory: Dr. F. Y. Henderson, C.B.E.

DR. F. Y. HENDERSON, who retires from the directorship of the Forest Products Research Laboratory, Princes Risborough, in October, was reader in timber technology at the Imperial College of Science and Technology. Initially interested in plant physiology, he took over Percy Groom's Sub-department of Technology of Woods and Fibres; the elementary text-book on wood technology which he wrote then proved specially popular with those in the timber trades. Henderson's tenure of the directorship marks a period in the history of the Forest Products Research Laboratory. He took over in 1945, when many relatively unknown woods were beginning to replace well-tried ones, and these needed investigation and assessment in relation to the traditional timbers; new and little-known insect and fungal pests claimed attention; plywood had developed enormously and the Laboratory played its part in research on such laminated wood. This work, and other aspects, like wood bending, machining and adhesives, might have been mentioned—it was Henderson's task to co-ordinate and guide. Moreover, the past few years have seen the enormous expansion of co-operative work with the Forestry Commission and the beginning of research on the pulping of home-grown timbers. The steady flow of important bulletins from the Laboratory emphasizes the volume of varied work carried out there since the War and, during this time, trade and private inquiries have occupied much of the time of the staff. Henderson's retirement marks the end, more or less, of this period. Relieved of most of the inquiry work, his colleagues have more time for the fundamental work for which the laboratory is well equipped. Moreover, reconstituted wood products, like hardboards and laminated wood, are replacing solid timber in many of its traditional uses and presenting their own special problems to the Laboratory.

Mr. H. Wooldridge, O.B.E.

MR. HENRY WOOLDRIDGE, who will succeed Dr. Henderson, has been a member since its formation of the Steering Committee of the Forest Products Research Laboratory. He has spent most of his career at the headquarters of the Department of Scientific and Industrial Research, where he is at present head of the Stations Division, and as such is responsible for liaison with all the Department's laboratories. Previously, he was deputy establishment officer in the Department, and before that he served two secretaries of the Department, Sir Frank Smith and Sir Edward Appleton, as private secretary. Mr. Wooldridge joined the Department in 1935 from the British Association for the Advancement of Science, of which he had been assistant secretary for five years. He was educated at Reading School and King's College, London.

Royal Society and Nuffield Foundation Commonwealth Bursaries

AWARDS under the Royal Society and Nuffield Foundation Commonwealth Bursaries Scheme have been made as follows: Dr. R. N. Brothers, senior lecturer in petrology, University of Auckland, to enable him to examine at the Imperial College, London, the application of crystalline and deformation theories for certain New Zealand ultrabasic rocks to a group of related rocks which occur in Great Britain, for a year from November 1959; Dr. J. W. Clark-Lewis, senior lecturer in organic chemistry, University of Adelaide, to enable him to continue a study of the chemistry of natural products, with particular reference to the stereochemistry of flavan derivatives, at the Imperial College, London, for seven months from October 1960; Dr. A. W. Crompton, director of the South African Museum, Cape Town, to enable him to study mammalian palæontology at Cambridge and the British Museum (Natural History) for four months during 1960; Dr. J. C. Gould, senior lecturer in bacteriology, University of Edinburgh, to enable him to extend his studies of staphylococcal infections and make comparative studies in an African community, in Nigeria, during May–September 1960; Prof. K. R. L. Hall, professor of psychology, University of Bristol, to enable him to carry out a field-study of behaviour and social organization of wild baboon groups in differing environments in South and South-West Africa, during July–September 1960; Dr. L. M. Jackman, lecturer in organic chemistry, Imperial College, London, to enable him to investigate hydrogen bonding in organic molecules by nuclear magnetic resonance spectroscopy, at Adelaide, during July–October 1960; Prof. D. O. Jordan, Angas professor of physical and inorganic chemistry, University of Adelaide, to assist him to visit University College, London, and the Royal Cancer Hospital, London, during February–September 1960, in order to study new techniques and particularly those concerned with the structure, behaviour and functioning of biological macro-molecules; Prof. H. Kleerekoper, professor of zoology, McMaster University, to assist him in completing during the summer of 1960 an investigation of bottom sediment cores of Lake Moneague, Jamaica, for which a bursary was awarded to him in April 1958; Mrs. Beryl E. March, instructor and research associate, Department of Poultry Science, University of British Columbia, to assist her to visit Cambridge during May–August 1960 in order to study methods of evaluating the nutritive value of fish meals; Dr. E. H. Mercer, of the Chester Beatty Research Institute, London, to enable him to carry out an electron microscopic study of cell division and differentiation in plants, at Sydney, during February–July 1960.

The Liverpool Geological Society

THROUGHOUT the first three days of 1960 the centenary celebrations of the Liverpool Geological Society, happily combined with the eighth Inter-University Geological Congress, attracted an enthusiastic gathering of more than two hundred geologists. The theme of the meetings was "Recent Advances in Geology", and each of ten lectures by speakers from British and foreign universities gave rise to spirited discussion. The Society has now published the text of these addresses in a volume of more than three hundred pages which is a fine contribution to geological literature (*Liverpool and Manchester Geol. J., Centenary Issue*: price 20*s*.). In it, T. S. Westoll reviews recent studies on the palæontology of fishes: G. Voll, in a sixty-page paper, summarizes the latest work in the field of petrofabrics; S. I. Tomkeieff describes sources of information on the geology of the U.S.S.R.; L. M. J. U. van Straaten writes on deltaic sedimentation; R. G. Mason on geophysical investigations of the sea floor; W. S. Mackenzie on experimental studies in petrology; W. C. Krumbein on numerical data in geology; C. F. Davidson on radiogeology and geochronology; and R. H. Cummings on advances in micropalæontology. An essay by W. E. Swinton tells the history of *Chirotherium*, the reptilian foot-print found in the Triassic rocks of Cheshire (long believed to be the hand-print of a mammal) which is reproduced on the seal and medal of the Society. Liverpool geologists have done well in sponsoring this readable review of present knowledge in some of the most active branches of earth science.

Uranium in South-West England

THE reconnaissance survey for uranium, carried out by the Atomic Energy Division of the Geological Survey during the past two years, is now virtually complete. Seventy new occurrences of uranium have been found during the course of operations which have included an aerial survey of Cornwall, Devon and much of Somerset, as well as surface mapping, excavating and drilling at promising localities. No deposit of economic importance has been discovered, and it is concluded that if any are present they are covered to such a depth with soil or overburden that they are unlikely to be detected by existing instruments. Although no deposit estimated to contain more than 5 tons of uranium oxide has been found as a result of this survey, the South-West of England uranium province has served for many years as an ideal testing ground for British geological electronic equipment, which is now used extensively throughout the world. The study of uranium occurrences in Cornwall and Devon, started in 1945, has played an important part in the discovery of uranium in the Commonwealth, while recent work has contributed considerably to the development of aeroradiometric surveying techniques.

Scientists and Engineers in American Industry

THE U.S. National Science Foundation has stated that American business firms employed about 780,000 scientists and engineers in January 1959. About 35 per cent of the total were engaged in engineering and scientific research and development activities. Scientists and engineers in the industry sectors included in the current survey are estimated to represent approximately two-thirds of the scientists and engineers employed in the United States. The other third are employed in government agencies, educational institutions, non-profit organizations, and self-employed. Industries employing the largest groups of scientists and engineers in January 1959 were aircraft and parts, electrical equipment, and chemicals and allied products. Each employed more than 75,000 scientists and engineers; combined, they employed one-third of the total number of scientists and engineers in private industry and approximately half the scientists and engineers engaged in the performance or administration of research-and-development activities in private industry. Each of two other industries employed more than 50,000 but less than 75,000 scientists and engineers: machinery (except electrical) and engineering and architectural services. Next in size, in terms of scientific and engineering employment, were construction, fabricated metal products and ordnance, primary metals, and motor-vehicles and equipment; each of these industry groups employed more than 30,000 but less than 50,000 scientists and engineers. Employment of scientists and engineers in private industry was about 4 per cent higher in January 1959 than in January 1958. Engineering employment increased from about 605,000 in January 1958 to 630,000 in January 1959. The number of scientists employed in private industry increased from about 145,000 in January 1958 to 150,000 in January 1959.

British Digital Computer Information Centre

The National Centre of Information on Automatic Programming of Digital Computers has been established by the Department of Mathematics of the Brighton Technical College in response to a recommendation of the first National Conference on Automatic Programming held in Brighton in April 1959. The purpose of the Centre is: to establish and maintain a comprehensive library of publications; to publish, in conjunction with Pergamon Press, Ltd., an annual review of automatic programming; to provide a 'clearing house' for information and inquiries on automatic programming; to organize small working conferences on particular aspects of the subject; and to maintain permanent contact with organizations in all other countries concerned with such matters. An advisory committee to assist the work of the Centre has been established and includes Dr. A. D. Booth (University of London), Mr. R. A. Brooker (University of Manchester), Mr. P. V. Ellis (International Computers and Tabulators, Ltd.), Dr. S. Gill (Ferranti, Ltd.), Dr. J. Howlett (United Kingdom Atomic Energy Authority), Dr. R. J. Ord-Smith (Standard Telephones and Cables, Ltd.), Mr. C. Robinson (English Electric Co., Ltd.), Mr. Christopher Strachey, Mr. A. E. Taylor (Remington Rand, Univac), Mr. J. H. Wensley (Computer Developments, Ltd.), and Mr. M. Woodger (Mathematics Division, National Physical Laboratory), together with the following as corresponding members: Dr. Grace Hopper (Remington Rand, U.S.A.), Dr. L. Luckaszewicz (Mathematical Machines Laboratory, Polish Academy of Sciences), and Prof. H. Rutishauer (Institute of Applied Mathematics, Zurich). Further information can be obtained from the organizing secretary, Mr. Richard Goodman, Department of Mathematics, Brighton Technical College.

Hospital Morbidity Statistics in Great Britain

A further contribution to the study of morbidity has recently been published by the General Register Office (The Registrar General's Statistical Review of England and Wales for the Year 1955. Supplement on Hospital In-Patient Statistics. Pp. vii+206. London: H.M. Stationery Office, 1959. 10*s.* 6*d.*). This volume summarizes the results of the hospital in-patient inquiry for 1955. This inquiry, which is steadily becoming more comprehensive, is expected to cover virtually the whole of the discharges of in-patients from hospitals in Britain by 1957; but in 1955 the hospitals that participated did so on a voluntary basis. Nevertheless the principal specialities seemed to be represented in close correspondence with the national figures, and the inquiry was fully representative of two hospital regions, East Anglia and Wales, for which special statistics are given. The statistical unit used in the inquiry is the discharge of an in-patient from a hospital, and the principal tables in the volume show the number of such discharges in various age and sex groups, and in different regions, by principal condition diagnosed. The number of deaths in hospital is shown separately, and there are data dealing with the duration of stay and the waiting period before admission to hospital. Special tabulations deal with children in hospital and maternity cases. Teaching and non-teaching hospitals are distinguished throughout. While hospital morbidity statistics are not a wholly reliable guide to morbidity, they can, as the report indicates, be of considerable assistance in the study of the mass aspects of disease, and the data will also be of considerable value to the hospital administrator. It is to be hoped that future tabulations will give information about the socio-economic status of the patients discharged. The appearance of this volume provides a valuable addition to our knowledge of the functioning of the National Health Service.

Transvaal Museum

After a lapse of four years it is fortunate that it was possible once again, during July 1959, to publish the *Bulletin of the Transvaal Museum*. A topical article on "Excavating Ape-Men", by Mr. J. T. Robinson, describes the cave limestone in which such remains are found. The grid system of excavation is described and all bone is treated with 'Glyptal' cement immediately after it is removed from the breccia. Afterwards a 10 per cent solution of acetic acid is used to free the bones from the surrounding matrix.

Musical Rites

Certain hard igneous rocks ring when struck and the note obtained depends on the dimensions of the rock itself. By careful shaping different notes can be obtained and a sort of 'glockenspiel' created. There is a decorated rock-shelter in Nigeria where such a 'glockenspiel' occurs to which parties come to be married, their faces being first carefully whitened. Dr. B. D. Malan describes just such a rock gong site in the Parys district of the Orange Free State in the *South African Journal of Science* (55, No. 2). It is well known that music plays a prominent part in the lives of many primitive folk; these gongs are very interesting and the resulting 'music' not at all unpleasant.

The Commonwealth Scholarship Commission

The Earl of Scarbrough has been appointed chairman of the Commonwealth Scholarship Commission in the United Kingdom. The Commission is to be set up under the Commonwealth Scholarships Act, 1959, recently passed by Parliament, to administer the Commonwealth Scholarship and Fellowship Plan in Britain. This Plan was worked out by the Commonwealth Education Conference at Oxford in July 1959 and creates a thousand new postgraduate awards throughout the Commonwealth. Of these, the United Kingdom will contribute five hundred scholarships and fellowships tenable in the United Kingdom at any one time.

University News: London

Dr. G. V. R. Born, research officer at the Nuffield Institute for Medical Research and departmental demonstrator in the Department of Pharmacology in the University of Oxford, has been appointed to the Vandervell chair of pharmacology tenable at the

Institute of Basic Medical Sciences. Dr. D. L. J. Bilbey, assistant professor of anatomy in the University of Toronto, and Dr. R. M. H. McMinn, lecturer in the University of Sheffield, have been appointed to readerships in anatomy tenable at King's College.

The title of professor of economics in the University of London has been conferred on Mr. B. S. Yamey in respect of his post at the London School of Economics and Political Science.

The title of reader in physics in the University of London has been conferred on Dr. D. M. MacKay in respect of his post at King's College.

Southampton

The Institution of Structural Engineers is to provide funds for the establishment of a readership in structural engineering in the Department of Civil Engineering of the University. The Institution will make available £2,000 per annum for seven years at the end of which period the Council will be responsible for the appointment.

Borough Polytechnic, London

The following appointments in the Borough Polytechnic have been made: G. Isserlis, senior lecturer in charge of the Division of Metal Science; Dr. D. B. Smith, senior lecturer in food technology; S. N. Houldsworth, lecturer in chemical engineering; Dr. B. E. Weller, lecturer in physical chemistry; Dr. H. W. Hykin, lecturer in mechanical engineering.

The Night Sky in February

Full moon occurs on Feb. 12d. 17h. 24m. U.T., and new moon on Feb. 26d. 18h. 24m. The following conjunctions with the Moon take place: Feb. 6d. 16h., Aldebaran 0·5° S.; Feb. 22d. 00h., Jupiter 5° S.; Feb. 23d. 04h., Saturn 4° S.; Feb. 24d. 07h., Mars 5° S.; Feb. 24d. 13h., Venus 4° S. In addition to these conjunctions with the Moon, Venus is in conjunction with Saturn on Feb. 7d. 11h., Venus being 0·2° N., and Venus with Mars on Feb. 17d. 03h., Venus being 1·1° N. Mercury can be seen as an evening star during the later part of the month, setting at 19h. 15m. on February 25. It reaches greatest eastern elongation on February 23. Venus is a morning star, rising about 2 hr. before the Sun at the beginning of the month. Later in the month it becomes too close to the Sun for observation. Mars is badly placed for observation, rising about an hour before the Sun. Jupiter is a morning star, rising at 5h. 00m., 4h. 20m. and 3h. 35m. on February 1, 15 and 29, respectively. It may be seen low in the south-east before sunrise; its stellar magnitude is −1·5. Saturn is also a morning star, but it is still too close to the Sun for satisfactory observation. Occultations of stars brighter than magnitude 6 are as follows, observations being made at Greenwich: Feb. 2d. 19h. 49·3m., μ Psc. (*D*); Feb. 6d. 14h. 26·0m., Aldebaran (*D*); Feb. 6d. 15h. 10·0m., Aldebaran (*R*); *D* and *R* refer to disappearance and reappearance, respectively.

"Salicylate and Potassium Fluxes of Rat Diaphragm"

We regret that certain printing errors appeared in the communication by Dr. J. A. Hicklin, published in *Nature* of Dec. 26, p. 2029. In the penultimate line of the second paragraph, the word 'active' should be deleted. In the third paragraph, the second sentence should read: "Contrary to expectation the principal effect of salicylate was consistently to accelerate the *efflux* of potassium-42", and the fourth sentence of the paragraph refers to estimates of *influx*.

Announcements

The Medical Research Council has recently established the following new research units: the Blood Coagulation Research Unit, by agreement with the United Oxford Hospitals, at the Churchill Hospital, Oxford, under the part-time direction of Dr. R. G. Macfarlane; and the Environmental Radiation Research Group under the honorary direction of Prof. F. W. Spiers in the Department of Medical Physics, University of Leeds.

Following the appointment of Captain R. A. Villiers as the first director of the Scientific Instrument Manufacturers' Association of Great Britain as from October 1, 1959, it has been announced that Mr. G. L. Knight has been appointed assistant director. Mr. Knight will operate under Captain Villiers in the field of information and external activities, and will be responsible for the collection and distribution of information for the Association.

A small travelling exhibit on rocket astronomy has been lent to the Science Museum, London, until the end of February by the United States Naval Headquarters. Pictures show the launching of rockets and photographs of the Sun taken from the rockets. An actual camera which has been recovered after use in a rocket is included in the exhibit, which is explained by a recorded talking commentary.

The Institute of Electrical Engineers has announced that the Faraday Lecture will be given by Prof. M. G. Say on February 17, at the Central Hall, Westminster, and will be entitled "Electrical Machines". Further information can be obtained from the Secretary of the Institute of Electrical Engineers, Savoy Place, London, W.C.2. Admission, which will be by ticket only, is free.

An all-day meeting of the Welsh Soils Discussion Group will be held on February 3 at University College, Aberystwyth. The programme will be introduced by Mr. J. R. Thom, director of forestry for Wales, and succeeding papers will deal with forestry and soils. Further information can be obtained from Mr. J. A. Taylor, Geography Department, University College, Aberystwyth.

A Symposium on "Basic Questions in the Structure of Languages" will be jointly sponsored by the American Mathematical Society and the Association for Symbolic Logic at the Hotel New Yorker in New York during April 14–16. At the same time and place the American Mathematical Society is also organizing a two-day symposium on "Stability Problems in Hydrodynamics". Further information can be obtained for both symposia from the Associate Secretary, American Mathematical Society, 190 Hope Street, Providence 6, Rhode Island.

A course of ten lectures entitled "Reactor Fuel Element Technology" will be given in the Department of Chemical Engineering at the Imperial College of Science and Technology on Thursday afternoons during February 11–March 17. This course is designed to lay emphasis on the industrial problems of reactor fuel-element technology. The speakers will be from the United Kingdom Atomic Energy Authority. Further information can be obtained from the Registrar, Imperial College of Science and Technology, London, S.W.7.

CENTENARY OF "THE ORIGIN OF SPECIES"

SOME BOTANICAL ASPECTS

THE Conference arranged by the Botanical Society of the British Isles to celebrate the centenary of the publication of the "Origin of Species" was held during November 27–29 and was well attended by members and visitors.

The President, in his introductory remarks, directed attention to the importance of Darwin's friendship with Hooker in the development of the botanical arguments in the "Origin".

Lady Barlow enlarged on this theme, among others, in the course of a most interesting and illuminating account of her grandfather's botanical activities. She pointed out that, though Darwin had benefited greatly from his taxonomic work on Cirripedia, his work on plants was entirely of an experimental kind and laid the foundations of a great deal of the subsequent development of these branches of botany.

Prof. T. W. Böcher (Institute of Plant-anatomy and Cytology, Copenhagen) traced the development of cytotaxonomy since Darwin's time. He pointed out that though Robert Brown discovered the nucleus when Darwin was twenty-two, and when he was only thirty-nine Hofmeister discovered chromosomes, what is now called cytotaxonomy was not born until this century. He regarded Rosenberg, who in 1909 demonstrated a polyploid series in *Drosera*, as the Father of this branch of botany. Rosenberg had many distinguished pupils, among them Tackholm and Heilborn, who together with Jørgensen, Tischler, Winge and, in Britain, Ruggles Gates, Blackburn and Heslop-Harrison placed the study on a firm footing in the 1920's. Among others who played an equally important part in this early development were Clausen, Hagerup, Langlet, Percival and Senjaninova. Meanwhile, in the U.S.S.R. under Navashin another school grew up more concerned with ideograms and karyotypes than with chromosome numbers.

Both these approaches, combined with the discovery of apomixis, permanent structural hybridity and allopolyploidy by later workers, have been extremely fruitful.

Not least among the benefits which botanists have derived from the growth of cytotaxonomy is the kind of information which should deter them from, as Prof. Bocher said, "attempting species-making in groups where any cutting up into described taxa will, at most, have only local or temporary importance".

The vital necessity of combining careful taxonomic work with cytological studies was pointed out and illustrated by reference to *Campanula rotundifolia* and *Triglochin maritima*, among others. *C. rotundifolia* is a variable complex consisting of rather local diploids, usually clearly distinguished from one another but resembling tetraploids growing in the same neighbourhood, which are themselves widespread and intergrading. The variation here seems to be mostly due to structural hybridity and has so far defied satisfactory taxonomic analysis. In the *Triglochin maritima* complex, on the other hand, there is a polyploid series ranging from $2n$ to $24n$ and at least some of the members of this series have distinct morphological characters and distributions, and correspond to previously described though not generally accepted species.

After surveying recent work on genera and families, Prof. Böcher pointed out that the names cytotaxonomy, experimental taxonomy, biosystematics or synthetic taxonomy are perhaps superfluous. It might be better just to use the word taxonomy, "presupposing that all taxonomists now and in the future to the best of their ability are cyto-geno-eco-choro-taxonomists".

Prof. W. H. Camp (Connecticut) in the course of an illuminating and entertaining hour described some of the ways in which evolution is occurring in the world to-day, illustrating his lecture with examples from his own experience. During the past century biologists had paid little more than lip-service to Darwin, and it was only recently that we had obtained a real insight into the mechanism of evolution.

His own approach to the problem had been *via* population genetics, which attempts, to begin with, to discard the classical species-concept and to study either populations in the field or samples of them in cultivation. From studies of this kind series of group concepts develop which are often, though not necessarily, identical with the traditional species, and at the same time a great deal of information is obtained about evolutionary processes which are going on now and often also about those which have taken place in the past. Knowledge of the cytology and breeding systems of the plants with which he is concerned, as well as the 'taxonomic eye', are essential for the accurate interpretation of the biosystematist's observations.

The variation in morphology and ecology in American species of *Fagus* and *Vaccinium* can be explained largely by the effects of Pleistocene changes in climate which gave the opportunity for introgression between a number of distinct but homoploid species, followed by selection for particular combinations of physiological and morphological characters adapted to the various habitats available in post-glacial times. Corroborative evidence for this is provided by the fossil remains of *Fagus* in various parts of the North Temperate regions and by the variation in kind and quantity of alkaloids in the bark of different *Cinchona* populations. In the *Vaccinium corymbosum* complex the situation is further complicated by the production of both auto- and allopolyploids.

Prof. Camp went on to demonstrate the extremely rapid changes which occur in *Viola* populations in response to variations in light-intensity, humidity and other habitat factors. This is a group in which morphological and physiological variation is very great and sterility barriers almost non-existent.

Attempts at subdivision into species seem to be meaningless as there is a continual evolution of new genotypes from chasmogamous flowers and, at the same time, the abundant production of seed from cleistogamous flowers allows of rapid increase of the best-adapted individuals in a population.

In conclusion, he suggested that evolution is perhaps best regarded not primarily as the production of new species but as the genetic ability of organisms to fill the multitudinous new habitats being constantly presented to them by the changing environment. Species may then be considered as 'ephemeral nodes in the genetic reticulum' within the space-time continuum.

Prof. D. H. Valentine (Durham) spoke about evolution in *Primula* section Vernales in which there are nine species, all with $2n = 22$, and all but two, *P. veris* and *P. lofthousei*, capable of crossing with fairly good meiotic pairing in the hybrids. Most species have long peduncles and erect capsules with dry seeds but in three with short peduncles the capsules lie on the ground. Of these *P. vulgaris* has sticky ant-dispersed seeds and occurs over quite a wide area, but the other two, with dry seeds, are very local endemics, the difference being perhaps attributable to the relative efficiency of the dispersal mechanisms.

It is possible to arrange the species of this section in a series based on the ease with which they can be crossed. This does not altogether agree with the morphological sequence, and, in the absence of almost all knowledge of the genetic mechanism and physiological basis of 'crossability', it is impossible to be certain which is nearer the phylogenetic truth. It is clear that evolution in this group has been gradual and continuous and is combined with geographical and ecological isolation rather than sterility barriers. In other sections of the genus, for example Farinosae, polyploidy has played an important part.

Dr. N. Hylander (Uppsala) gave an account of some trends in modern research work on Scandinavian plants. He pointed out that a number of species previously unknown in Scandinavia had been discovered, even in the past decade, a remarkable thing in a region which has for long possessed an unusually high concentration of botanists. The possibility of perglacial survival has been the subject of many investigations, and, Dr. Hylander thought, had perhaps been rather overdone. So many species have been added to the list of survivors that the supposed nunataks now gave the impression of having been overpopulated. Endemism in the *Papaver radicatum* group, he suggested, might after all be of relatively recent origin. The ecotype concept of Turesson is an important Scandinavian contribution to botany and one which, combined with cytological work, has occupied the attention of many workers.

Variation within agamospecies has been observed in a number of plants and the basis of it investigated by Sørensen and Gudjonsson in triploids of the *Vulgata* group of *Taraxacum*. Here it has been shown that loss of one, or rarely two, chromosomes is responsible for most of the variations, and that *gigas* forms arise through polyploidy. Detailed study of the taxonomy and the method and speed of dispersal of microspecies has yielded much information about plant migrations and the age of different floristic elements.

Prof. I. Manton (Leeds), before proceeding to discuss some aspects of evolution in Pteridophyta, said how much she owed to the stimulation and instruction she had received from both Rosenberg and Prof. W. H. Lang. She then outlined some of the main aspects of the cytology of the group, pointing out that, since high basic numbers and polyploidy were frequent, it was important for cytologists to be well acquainted with the twenty-nine times table. In some tropical species of *Ophioglossum* diploid numbers greater than 1,000 occur and it is difficult to imagine the mechanism by which pairing is achieved at meiosis.

Allopolyploidy appears to be the chief means by which species have been formed, at least among Polypodiaceae in the broad sense. About 50 per cent of British ferns are known to be allopolyploids, one parent of which is often known; the other parent is sometimes to be found in N. America and most of those at present unknown will probably be found in temperate Asia when that region is better explored botanically.

From a fairly exhaustive study of the cytology of Pteridophyta in Britain, Madeira, Ceylon, Malaya and Tropical Africa it is evident that in this group polyploidy is not a response to cold, as has been suggested for angiosperms. In Ceylon and Tropical Africa 60 per cent of the species so far investigated are polyploids.

Natural hybrids occur among the ferns far more frequently than is generally recognized, but the F_1 generation is often so like one parent that they pass unnoticed. Prof. Manton estimated the number of living species of ferns at approximately ten thousand and between them at the very least five thousand hybrids occur. It is thus evident that ample opportunities exist for continued evolution through allopolyploidy.

Dr. G. D. H. Bell (Plant Breeding Institute, Cambridge) demonstrated how 'pure' cytotaxonomy was being applied to the improvement of crop plants. He made it clear that the highest taxonomic skill combined with a good knowledge of cytology and genetics, and adequate facilities, were essential for this work and that so far, even with *Triticum*, no more than a small beginning had been made.

The collection of wild relatives of crop plants from centres of genetic variation followed by a thorough study of the genetics, cytology and taxonomy of the material provide the basic information, which in *Triticum* has led to a good understanding of the genomes of the cultivated hexaploids. New amphidiploids produced, with the aid of colchicine, from crosses between *Aegilops ovata* and $6n$ wheats have enabled increased genetic variability to be introduced into this important crop plant. Resistance to *Erysiphe graminis*, for example, can be brought in from the *Aegilops* parent and promises to be valuable. A technique has recently been worked out for introducing one or two *Secale* chromosomes into hexaploid wheat through suitable hybridizations, thus giving an entirely new source of variation which may prove of practical importance.

Dr. Bell pointed out that not only was taxonomy *sensu* Bocher the basis of plant-breeding but also that the benefits were reciprocal; indeed, work of this kind could go far towards solving the problem of world food supplies.

An interesting series of exhibits illustrating recent research on the subjects of Darwin's botanical works was on view and directed attention to the variety of lines in which he was the pioneer. It is perhaps not generally known, for example, that he was the first person to use coleoptiles in the experimental investigation of phototropism.

A highly successful and enjoyable meeting concluded with a conversazione and a visit to Down House.

A full account of the Conference will be published by the Society. T. G. TUTIN

UNIVERSITY STUDENTS AND THEIR ACADEMIC ORIGINS

WITHIN the past ten years there have been several valuable studies of student performance at universities in the United Kingdom, some concerned with 'wastage', or failure to complete a course of study; some with degree of success achieved by those who did not fall by the wayside; and some (of which the survey under notice is an example) with both. A variety of factors make it difficult to compare the results of such investigations. First, they usually relate to one particular university or college, where conditions affecting initial selection of students and their subsequent academic progress may differ materially from those obtaining elsewhere. Secondly, they are often based on the intake of a particular year (differing from the year chosen in other surveys), and year-to-year fluctuations in the quality of students are believed to be considerable, at least within one college, faculty or department. Thirdly, to serve the purpose of the inquiry, success or failure is analysed in terms of a limited number of characteristics, which vary from one survey to another.

The limitations imposed by these and other factors are well brought out if the survey relating to the Queen's University of Belfast (An Audit of Academic Performance: a Study of the Prediction of Academic Success in the Queen's University of Belfast. By Michael Forster. Pp. 75. Belfast: Queen's University, 1959. 7*s.* 6*d*.) is put side-by-side with another very recent one, undertaken by the Cambridge University Sociological Society (Academic Careers of Entry of 1955). There is no need to emphasize the importance of the different setting provided by the Queen's University of Belfast as compared with the University of Cambridge for a study of academic performance. In the Queen's University case the students concerned are those who entered in the four-year period 1946–49; for Cambridge, the intake of one session, 1955–56, was taken. For the purpose of the Belfast inquiry, degree of academic success was analysed in terms of four main possible associated factors: performance in the Northern Ireland Senior Certificate Examination (the main mode of entry), age at entry, whether educated at a Belfast or a non-Belfast school, and whether a scholarship holder or not. In the Cambridge inquiry the factors selected were two in number. The first was type of secondary school attended, and here the main classification separated Headmasters' Conference, Direct Grant, Grammar and other schools from each other (but did not, unfortunately, distinguish between predominantly boarding and predominantly day schools within these categories). The second analysis involved comparing the performance of entrance scholars and exhibitioners with that of commoners.

Although few comparisons can usefully be made between two studies with such very different settings and purposes, this does not, of course, imply that they were not worth undertaking. Their value lies, to begin with, in providing a factual basis for discussion of the topics with which, individually, they were designed to deal. Is the examination, on the basis of which entrance to the Queen's University is largely determined, a satisfactory index of probable future performance? Are Cambridge colleges admitting too high a proportion of entrants from certain types of school, at the expense, perhaps, of other entrants more likely to show good academic results? Another obvious value of these surveys is that, when further investigations are made in the same universities for later years, it will be possible to identify some of the changes that have taken place. Again, each new survey makes some contribution to the methodology of work in this particular field. Finally, where a number of inquiries relate to the intake of different universities in the same year, there is at least the possibility of fruitful inter-university comparison. The Cambridge survey, for example, has taken the year used in the study of admissions undertaken by me for the Committee of Vice-Chancellors and Principals, and this is also the basis on which Prof. D. V. Glass and his colleagues are conducting a large-scale follow-up inquiry which should throw considerable further light on the social factors influencing academic success among students at British universities.

R. K. KELSALL

PLANT–ANIMAL FOOD CYCLES IN THE SEA

MICROSCOPIC phytoplankton floating passively in the sea have proved a suitable field for the techniques of population dynamics. Gordon Riley and others have shown that when populations of plants, and also the herbivores that graze on them, are considered in terms of their content of organic carbon, it is possible to predict roughly the varying quantities in different areas in terms of the changes in the physical environment. The problem, however, from the point of view of fisheries studies, is that only one part of this generalized organic matter may be relevant. Thus, herring will feed on copepods but not on salps, both of which graze on the phytoplankton. In this way an area of high primary production is a necessary but not a sufficient condition for a commercial fishery.

A paper by Dr. D. H. Cushing, of the Fishery Laboratory at Lowestoft, is of interest because in a study of the organic production in the southern North Sea he has introduced details of the species of plants and animals involved ("On the Nature of Production in the Sea". Fish Invest., Lond., Ser. II, Vol. 22, No. 6. Pp. iii+40. London: H.M. Stationery Office, 1959. 10*s.* net). In place of the abstract idea of quantities of animal carbon eating quantities of plant carbon, he has developed an 'encounter' theory of grazing relating the speed of movement of copepods and the density of plant cells. This gives grazing-rates which are realistic in terms of the experimentally determined food requirements of the animals and the rates of depletion of plants in natural environments. His other innovation is to compute the growth of the *Calanus* populations, which are the dominant herbivores in the area studied, in terms of: (1) relations between plant density and egg laying; (2) the times taken to pass through the naupliar and copepodite stages; (3) probable mortalities at the various stages. His theoretical model of the production during the spring outburst of phytoplankton then gives results in terms of the numbers of *Calanus* (and similarly other copepods) that will be present at different times. The usefulness of this analysis has been shown in Dr. Cushing's previous work, where his results suggested that the aggregation of herring shoals on *Calanus* patches and their subsequent disengagement depended on a threshold concentration of *Calanus* in the patches ("Production and a Pelagic Fishery." Fish. Invest., Lond. Ser. II, Vol. 18, No. 7. Pp. 112. London: H.M. Stationery Office, 1955. 12*s*. 6*d*. net).

These detailed advantages, however, have certain drawbacks. Analyses in terms of particular species do not permit the easy comparison with other areas which is an important feature of the more abstract methods. For example, by conversion from plant cell-volume, Dr. Cushing gets a value for basic production during the spring. This value is only about one-quarter of those usually calculated for comparable areas, but it is difficult to see reasons for this divergence.

Another disadvantage of Dr. Cushing's model is that the possible effects on the plants of low nutrient concentrations are not included. Theoretically, this factor is used to make the more 'conventional' models stable; practically, it allows them to deal not only with the spring outburst when nutrients are comparatively plentiful, but also with summer conditions when they are scarce. Dr. Cushing points out, however, that although nutrient supply finally determines the quantity of production, this supply may depend not merely on the rate of uptake by the plants but also on the rate of return of inorganic nutrients to the water through herbivore excretion—a factor about which we know far too little. This could be particularly important in tropical oceanic areas where nutrients are at very low concentrations and where the high water temperatures may accelerate the biological re-cycling. Dr. Cushing's suggestion that production in these areas may be higher than formerly supposed is borne out by recent evidence, given at the International Oceanographic Congress in New York in September by Menzel and Ryther, that a significant outburst of production can occur in the northern Sargasso when nutrients are extremely low. Thus, production may depend on a balance between the physical and biological rates at which nutrients are made available to the plants. In the richer temperate and arctic areas, production may be mainly controlled by the supply of nutrients from the deeper waters, but it is possible that in tropical areas the rate of production is largely independent of such features of the physical environment and depends on how fast the plant-herbivore communities can re-cycle their nutrients.

Dr. Cushing, by his individual approach to these questions, has shown up some of the failings of the abstract methods, and particularly the need to make use of information about species composition and behaviour.

J. H. STEELE

IMMUNITY OF MICE SURVIVING SYSTEMIC LEUKÆMIA (*L*1210) TO ANTIFOLIC RESISTANT VARIANTS OF THE DISEASE

By DR. ABRAHAM GOLDIN, STEWART R. HUMPHREYS, GERALD O. CHAPMAN, DR. MICHAEL A. CHIRIGOS and JOHN M. VENDITTI

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TWO of the halogenated derivatives of amethopterin, 3′5′-dichloroamethopterin and 3′-bromo-5′-chloroamethopterin, have been demonstrated to be markedly effective in the treatment of the transplantable acute lymphoid leukæmia *L*1210 in mice[1-4]. Employing these derivatives of amethopterin, it was possible to obtain extensive survival time and an appreciable number of survivors of systemic leukæmia *L*1210, even when treatment was initiated only several days prior to anticipated day of death.

With a relatively high level of inoculum (about 1,000,000 cells/mouse) injected subcutaneously, the *CDBA* mice employed in the above studies $(BALB/c\ An \times DBA/2J)F_1$ male mice succumbed characteristically in 10–12 days with systemic leukæmia, and a local tumour at the site of leukæmic inoculation (10–12 mm. in diameter). On treatment of advanced leukæmia with the dihalogenated derivatives of amethopterin, the local tumour at the site of leukæmic inoculation usually regressed after several treatments, and survival times of 90 days and beyond were not uncommon[4].

The mice that survived systemic leukæmia following treatment with the halogenated derivatives of amethopterin showed immunity on reinoculation of the leukæmia[4,5]. Many of the mice appeared to be highly refractory, and the leukæmic re-inoculum showed no evidence of growth. Other mice showed partial immunity. In the latter case, the re-inoculum grew slowly, but progressively, and the animals

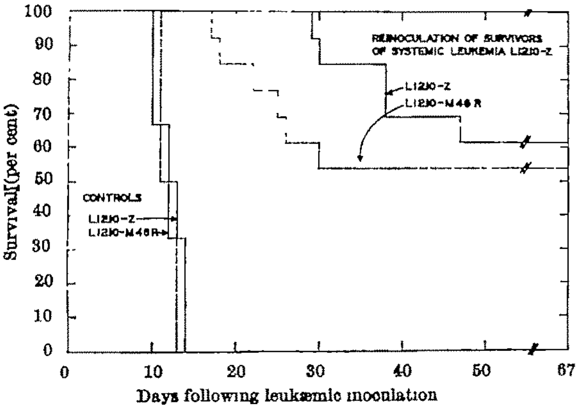

Fig. 1. Inoculation of the sensitive leukæmia (*L*1210*Z*) and an amethopterin-resistant subline (*L*1210–*M*46*R*) into *CDBA* mice surviving systemic leukæmia *L*1210 following treatment with 3′5′-dichloroamethopterin and 3′-bromo-5′-chloroamethopterin. Following treatment of leukæmic (*L*1210*Z*) mice with 3′5′-dichloroamethopterin and with 3′-bromo-5′-chloroamethopterin from day 7–90 there were 45/290 survivors on day 160. The mice were re-inoculated with *L*1210*Z* at that time, and there were 26/45 survivors on day 218 (ref. 5). These 26 mice were employed in the current study. Re-inoculated animals, 13 per group; controls, 6 mice per group, including the animals given the experimental and 1/10 of the experimental inoculum. Day 0 equals day 218 following the initial inoculation of leukæmia *L*1210. *L*1210*Z* Gen. 242; *L*1210–*M*46*R* Gen. 56. Approximately 500,000 cells per mouse, subcutaneously

eventually died, showing a rather extensive increase in survival time as compared with controls.

Similarly, the immune response to leukæmia *L*1210 has been elicited in 'out of strain' mice without the need for therapy[5]. When leukæmia *L*1210 was introduced subcutaneously into *BALB*/*c An* mice it grew at the site of inoculation. In some instances the growth was progressive, resulting in the death of the *BALB*/*c An* mice. In most instances the tumour regressed following the initial growth, and the mice were immune to re-inoculation of the disease.

The current study was undertaken to determine whether *CDBA* mice, apparently cured and immune to leukæmia *L*1210 as the result of therapy with the halogenated derivatives of amethopterin, or *BALB*/*c* mice, made immune to leukæmia *L*1210 following spontaneous regression of the tumour, would also be immune to antifolic resistant variant sub-lines of leukæmia *L*1210. For this study two antifolic resistant variants were employed: (*a*) An amethopterin-resistant variant of leukæmia *L*1210 (*L*1210–*M*46*R*). This variant of leukæmia *L*1210 appeared in response to therapy with amethopterin over several tumour generations[6]. *L*1210–*M*46*R* is resistant to therapy with amethopterin and shows a high degree of cross-resistance to the halogenated derivatives of amethopterin[6]. (*b*) The variant *L*1210–*M*66–3*R* was developed by therapy with 3′5′-dichloroamethopterin in serial transfer of leukæmia *L*1210. It has been demonstrated to be resistant to treatment with 3′5′-dichloroamethopterin over a wide range of doses[7].

The results of re-inoculation of drug-treated *CDBA* survivors of systemic leukæmia (*L*1210) with leukæmia *L*1210 and the resistant variant *L*1210–*M*46*R* are shown in Fig. 1. Treatment with 3′5′-dichloroamethopterin and 3′-bromo-5′-chloroamethopterin had been administered daily from the 7–90 day following leukæmic inoculation and the second inoculation with leukæmia *L*1210*Z* of the survivors had been on day 160[4,5]. The third inoculation (Fig. 1) employing *L*1210*Z* and *L*1210–*M*46*R* was on day 218. Fig. 1 shows that the mice with systemic leukæmia *L*1210, rendered immune by extensive treatment with the halogenated derivatives 3′5′-dichloroamethopterin and 3′-bromo-5′-chloroamethopterin, were immune, not only to the inoculation of the *L*1210 sensitive tumour, but were also immune to the *M*46*R* resistant variant of leukæmia *L*1210.

With both the *L*1210*Z* and the amethopterin-resistant variant *M*46*R* there were a considerable number of animals alive at 67 days following inoculation (285 days since the first inoculation of the leukæmic cells) (Fig. 1). At 67 days there were 8 out of 13 mice alive that had been re-inoculated with the sensitive tumour and 7 out of 13 mice alive that had received the amethopterin-resistant *M*46*R* tumour. The animals that did die following re-inoculation with *L*1210 or the *M*46*R* variant showed a marked increase in survival-time as compared with the controls inoculated for the first time. In general, as had been observed earlier for leukæmia *L*1210 in immune mice[4], they succumbed with large local tumours at the site of inoculation. The control mice for both the sensitive and resistant sub-lines had median survival times of 12 days. (The control mice employed in this experiment were somewhat younger than the immune mice. However, as indicated earlier[4], the immune phenomenon could not be attributed to an age factor. In more recent studies, this has been re-checked for both leukæmia *L*1210*Z* and the *L*1210–*M*46*R* variant.) For both the sensitive and resistant sub-line of leukæmia *L*1210 the inoculum-level employed was well above that required to give 100 per cent takes in control mice inoculated with tumour for the first time. With the *L*1210*Z* the experimental inoculum was 10 times greater than that giving 100 per cent takes in controls, while with the *L*1210–*M*46*R* it was 100-fold greater than that required to give 100 per cent takes in controls.

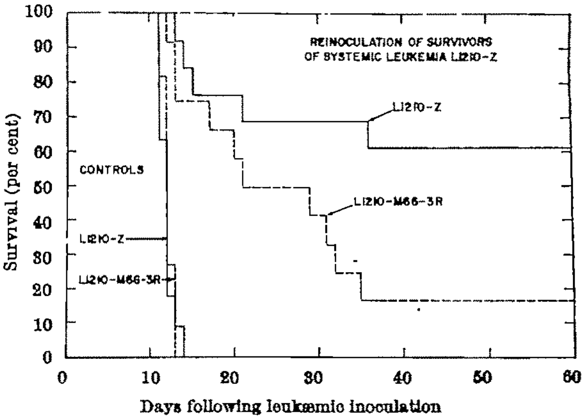

Fig. 2. Inoculation of the sensitive leukæmia (*L*1210*Z*) and a 3′5′-dichloroamethopterin-resistant subline (*L*1210–*M*66–3*R*) into *CDBA* mice. The re-inoculated mice had survived systemic leukæmia following treatment with 3′5′-dichloroamethopterin. Thirteen mice were re-inoculated with leukæmia *L*1210*Z* and 12 mice were re-inoculated with *L*1210–*M*66–3*R*. These mice were survivors of several experiments. All had been treated with 75 mgm./kgm. 3′5′-dichloroamethopterin starting at 7 days following leukæmic inoculation, and with one exception treatment was to the 50–80th day. Three of the mice re-inoculated with *L*1210 and four of the mice inoculated with *L*1210–*M*66–3*R* had already been shown to be immune to *L*1210. The remainder of the long-term survivors were re-inoculated for the first time. Controls, 11 mice/group of age comparable to that employed in the experiments. *L*1210*Z* and *M*66–3*R* inocula approximately 500,000 cells per mouse, subcutaneously. The inoculum concentration for both tumours was 100 times greater than that required to give 100 per cent takes in controls. *L*1210*Z* generation No. 267. *L*1210–*M*66–3*R* generation No. 20

Table 1. INOCULATION OF LEUKÆMIA *L*1210*Z* AND AN AMETHOPTERIN RESISTANT SUBLINE (*L*1210–*M*46*R*) INTO *BALB/c An* MICE IMMUNIZED AGAINST THE SENSITIVE LINE (*L*1210*Z*)

	First inoculation (*L*1210*Z*) Day 0			Second inoculation (*L*1210*Z*) Day 38			Third inoculation (*M*46*R*) Day 60		
	Takes / Total	Max. tumour diam. Av. mm.	Survivors / Total Day 38	Takes / Total	Max. tumour diam. Av. mm.	Survivors / Total Day 60	Takes / Total	Max. tumour diam. Av. mm.	Survivors / Total Day 128
BALB/c An	10/10	12 1	6/10	0/6	0	6/6	0/6	0	6/6
BALB/c An				10/10	10 5	9/10	0/9	0	9/9
BALB/c An							10/10	12	6/10
CDBA							12/12	10	0/12

Approximately 25 mgm. spleen inoculated per mouse (see ref. 5).

Fig. 2 summarizes results obtained when the variant *L*1210–*M*66–3*R* was utilized in a similar experiment. *CDBA* survivors of systemic leukæmia *L*1210*Z* exhibited immunity not only to subsequent inoculation of *L*1012*Z*, but also to the variant *L*1210–*M*66–3*R* resistant to 3′5′-dichloroamethopterin. Without any further therapy immune mice re-inoculated with each of these tumours showed extensive survival-time as compared with controls inoculated with either tumour for the first time.

It is of interest to note that the immune response appeared to be more extensive in mice re-inoculated with *L*1210*Z* as compared with *L*1210–*M*66–3*R*. There were 8 out of 13 sixty-day survivors on re-inoculation with *L*1210*Z* and 2 out of 12 sixty-day survivors following inoculation with *L*1210–*M*66–3*R*. By the same token the immunity to rechallenge with *L*1210*Z* appeared to be somewhat more extensive than to challenge with the *L*1210–*M*46*R* (Fig. 1). The nature of these differences is under further investigation.

Similarly, mice of strain *BALB/c An* immunized against leukæmia *L*1210, without drug therapy, were also immune to subsequent inoculation of the resistant variants *L*1210–*M*46*R* and *L*1210–*M*66–3*R*. This is illustrated for the *M*46*R* variant in Table 1. Group 1 had received the sensitive *L*1210*Z* as the first implant[5]. In 4 out of 10 mice the tumour grew progressively and the animals succumbed. In the remaining 6 mice the tumour regressed and the animals survived. On the second inoculation with leukæmia *L*1210 the tumour failed to grow. The third inoculation was with *M*46*R* and the tumour did not take.

In group 2 (Table 1) the *L*1210*Z* had been inoculated once into the *BALB/c An* mice. The tumour grew locally and then regressed in all but one mouse. The nine survivors were inoculated with the *L*1210–*M*46*R* resistant variant and showed no evidence of tumour growth. In contrast, *BALB/c An* mice receiving the *M*46*R* variant for the first time (group 3) developed large local tumours at the site of inoculation, averaging 12 mm. in diameter. Four out of 10 mice succumbed to progressive tumour growth. In the remaining 6 animals the tumour regressed.

It is noteworthy that the *CDBA* mice and leukæmia *L*1210 appear to be highly compatible. The immune response has not been observed in non-treated leukæmic *CDBA* mice. In this hybrid the leukæmia has never been observed to undergo spontaneous regression, and survivors of inoculum-levels too low to produce leukæmia have invariably been susceptible to re-inoculation.

In all the above experiments the source of inoculum was a suspension of leukæmic spleen taken from *DBA*/2 mice, the strain of origin[8]. It is of interest to point out that attempts to immunize *CDBA* mice against *L*1210*Z* by pre-inoculation of *DBA*/2 spleen from normal mice have been unsuccessful[6]. The immune response in *CDBA* mice has been observed only following successful therapy of leukæmia with the halogenated derivatives. Results were obtained suggesting that the immune response is not readily demonstrable unless successful therapy is initiated after the disease has become systemic[5].

On the other hand, transplantation of *DBA* spleen from normal mice into out-of-strain mice has been demonstrated to immunize mice against subsequent inoculation of *L*1210 or the antifolic variants[7].

Since the leukæmic mice treated successfully with the dihalogenated derivatives of amethopterin developed immunity to the disease, it has been suggested that this immunity may contribute to the success of the drugs in chemotherapy[4]. Conceivably, chemotherapy may so alter or influence tumour cells that homograft characteristics are acquired or unmasked, thereby facilitating further therapy[5,9]. This would raise the question as to whether the origin of resistance to antifolic therapy is attributable to resistance to the drug *per se*, or to resistance to a therapeutically induced immune mechanism.

The current studies provide a separation between tumour cell resistance to therapy as opposed to tumour cell resistance to host immunity. The two variants of leukæmia *L*1210, *M*46*R*, and *M*66–3*R*, are resistant to antifolic therapy but are inhibited by the induced immunity of the host. Thus, although the immune mechanism may contribute to therapy, resistance to therapy does not necessarily imply abrogation of the effectiveness of the immune response. Interrelationships of chemotherapeutic and immunological phenomena in host-tumour systems are under further study. Elucidation of such phenomena may contribute to our knowledge of both transplantation and chemotherapy.

We wish to thank Gerome McDowell for capable technical assistance.

[1] Goldin, A., Venditti, J. M., Humphreys, S. R., and Mantel, N., *J. Nat. Cancer Inst.*, 19, 1133 (1957).

[2] Goldin, A., Venditti, J. M., Humphreys, S. R., Shuster, L., Darrow, R. A., and Mantel, N., Abstracts 133rd Meeting Amer. Chem., Soc., 25–26*M*, San Francisco, California (April 1958).

[3] Goldin, A., Venditti, J. M., Humphreys, S. R., Shuster, L., Darrow, R. A., and Mantel, N., Acta Unio Internat. Contra Cancrum (in the press).

[4] Goldin, A., Humphreys, S. R., Venditti, J. M., and Mantel, N., *J. Nat. Cancer Inst*, 22, 811 (1959).

[5] Goldin, A., and Humphreys, S. R., *J. Nat. Cancer Inst.*, 24, No. 2 (Feb. 1960).

[6] Humphreys, S. R., and Goldin, A., *J. Nat. Cancer Inst*, 23 (Oct. 1959).

[7] Humphreys, S. R., Chirigos, M., and Goldin, A. (unpublished observations).

[8] Law, L. W., Dunn, T. B., Boyle, P. J., and Miller, J. H., *J. Nat Cancer Inst*, 10, 179 (1949).

[9] Klein, G., *Cancer Res.*, 19, 343 (1959).

REDUCTION OF INTESTINAL ABSORPTION BY A SYNTHETIC CHEMICAL

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A SYNTHETIC bactericidal substance, trimethylhexadecylammonium stearate, was submitted to a study of pharmacological properties prior to a proposed trial in certain human alimentary infections. Preliminary studies showed loss of weight, which was first believed to be due to reduced food intake. Daily measurement of the food residue of control and treated groups showed no reduction in the food consumption of the latter, however, until a very late stage when the animals became moribund.

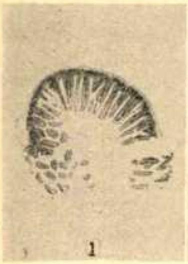
The animals used in these experiments were mainly adult male mice from non-interbred *O* stock. The water-insoluble trimethylhexadecylammonium stearate was incorporated in diet 41 (powder form), 5 gm. of which was given to each mouse per day. The concentration of the drug and the loss in weight after one month, or in the case of the three higher doses, at the time of death, are shown in Table 1. Analysis of variance gave $P \leqslant 0 \cdot 001$ for linear regression and for the variance between doses. Deviation from regression was not significant.

The histological appearances in mice receiving 0·4 and 0·5 per cent of the drug were compatible with the varying degrees of damage found in starved mice. In addition, most of the animals in these two groups showed at the time of death coffee-ground hæmorrhages in a specific area extending from the pyloric portion of the stomach to the ileum. This was invariably associated with damage to the intestinal villi, which involved primarily the epithelial lining. No hæmorrhages were noted in groups receiving smaller concentrations of the drug, though the animals still showed the appearances typical of inanition with total absence of depot fat and reduction in the weight of the testes and seminal vesicles ($P < 0 \cdot 001$). Animals receiving 0·1 per cent trimethylhexadecylammonium stearate died after 2–4 months of treatment, but some survived longer. The fæces were well formed in all groups, and the loss of weight could not, therefore, be attributed to intestinal hurry due to irritation. A few of the animals given 0·05 per cent of the drug for three months or more showed marked hypertrophy and hyperplasia of the duodenum and jejunum (Figs. 1 and 2).

Adult mice receiving 0·02 per cent trimethylhexadecylammonium stearate for six months did not differ from control mice either in weight or in general appearance. The rate of growth of immature mice given this concentration of the drug for periods of a fortnight at a time, in alternation with equal periods on diet 41, was depressed. The average fortnightly gain in weight per mouse was 2·5 gm. on the drug, as compared with 3·4 gm. for the control group ($P < 0 \cdot 001$).

Table 1. WEIGHT LOSS AND SURVIVAL PERIOD OF MICE ON DIFFERENT CONCENTRATIONS OF TRIMETHYLHEXADECYLAMMONIUM STEARATE IN THE DIET

Conc. of drug in diet (gm. per cent)	No and sex of mice	Original weight (gm.)	Weight at 1 month (gm.)	Difference (gm.)	Survival period	Average change in weight (gm.)
0·0						
a	6 m.	146·1	147·0	+0·9		
b	6 m.	140·4	140·5	+0·1		
c	6 m.	148·9	141·0	−7·9		
d	6 f.	128·6	125·7	−2·9		−0·41
0·025						
a	6 m	139·3	141·7	+2·4		
b	6 m.	148·9	147·5	−1·4		
c	6 f.	128·1	127·9	−0·2		+0·04
0·05						
a	6 m.	145·8	144·0	−1·8	3 months or over	
b	6 m.	138·9	141·2	+2·3		
c	6 m.	149·6	150·0	+0·4		
d	6 m.	148·3	147·0	−1·3		
e	6 f.	128·4	125·0	−3·4		−0·12
0·1						
a	6 m.	146·0	138·0	−8·0	2–5 months	
b	6 m.	140·0	126·7	−13·3		
c	6 m.	148·5	138·0	−10·5		
d	6 f.	128·5	118·2	−10·3		−1·75
0·2						
a	6 m.	144·5	120·5	−24·0	17–35 days	
b	6 m.	140·2	114·5	−25·7		
c	6 m.	148·9	127·0	−22·9		
d	6 f.	126·9	117·8	−11·1		−3·49
0·4						
a	6 m	153·0	118·0	−38·3	11–20 days	
b	6 f.	126·8	103·1	−23·7		−5·16
0·5						
a	6 m.	146·0	106·8	−39·2	3–12 days	
b	6 m.	134·4	110·7	−25·9		
c	6 m.	149·5	120·0	−29·6		
d	2 m.	54·5	39·5	−14·9		−5·48



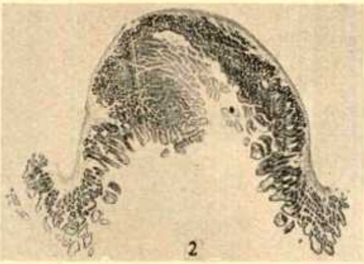


Fig. 1. Control: jejunum of normal mouse

Fig. 2. Jejunum of mouse kept on 0·05 per cent trimethylhexadecylammonium stearate in diet 41 for 3 months

A suspension of trimethylhexadecylammonium stearate in 0·5 per cent 'Tween 80' was administered to mice by stomach tube in doses of 25, 10, 5 and 2·5 mgm. daily. Given in this manner, the drug proved 3–5 times more effective than in diet 41. The animals died within 3, 7 and 9 days, and showed similar loss of weight and histopathological findings after the first three doses. Reduction of appetite became evident only in the terminal stages. With the latter dose, there was loss of weight comparable with that obtained on 0·1 per cent in the diet. The mice receiving this dose were alive and well after 10 weeks of treatment, though slimmer than a control group receiving 'Tween 80' only by the same route.

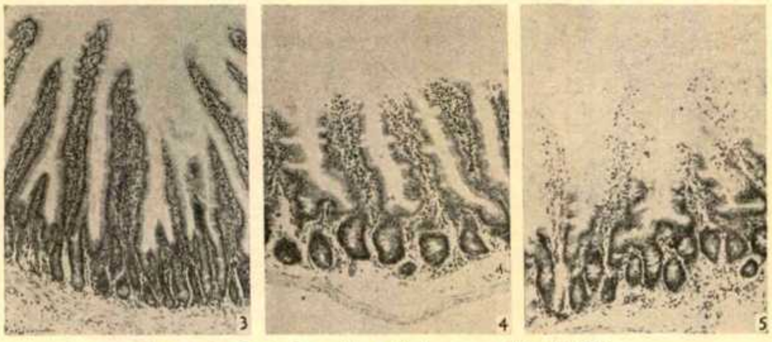
Trimethylhexadecylammonium stearate was more toxic when given subcutaneously in the form of the 'Tween' suspension. The *LD*50 was of the order of 0·6 gm./kgm. body-weight, a dose smaller than that received in a single day in a diet containing 0·5 per cent of the drug. After a dose of 1·0 gm./kgm., all the mice died within 24 hr., and 18/20 of these animals showed the characteristic intestinal hæmorrhages due to damage of the mucosa and villi in the pyloric area, duodenum and jejunum (Figs. 3–8). Single doses of 0·25 gm./kgm. did not cause death, but the animals continued to lose weight for some 10 days, and the difference in weight from a control group at one week was highly significant ($P < 0{\cdot}001$).

Administration via the intraperitoneal route proved more toxic still. The *LD*50 was of the order of 0·05 gm./kgm. for C_3H mice and 0·08 gm./kgm. for ordinary stock mice. Post-mortem usually showed marked dilatation of the stomach and intestine in addition to the previously described findings. Doses of 0·025–0·05 gm./kgm. given weekly for 10 weeks to eight mice on unrestricted diet resulted in gain of weight of 0·5 gm. as compared with 2·5 gm. per mouse in the control group.

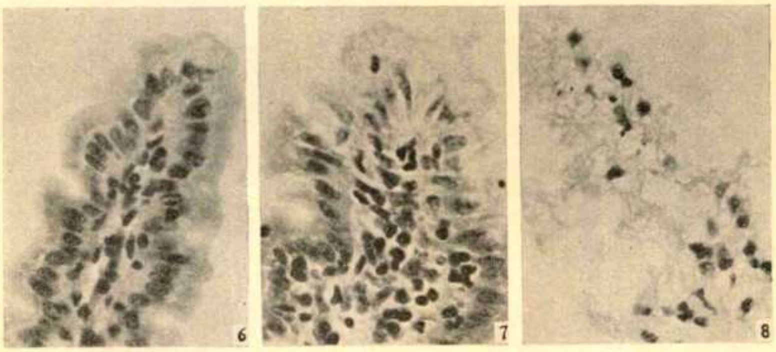
Animals allowed to recover after losing a fifth to a quarter of their body-weight (average loss = 4 gm.), either by continued administration of the drug in the diet (12 mice) or by a single subcutaneous administration (12 mice), were restored to normal weight when put on unrestricted normal diet. There was no obvious injurious effect on health up to three months after the end of the experiment, and the animals were indistinguishable from those in the control group.

The above results suggest that trimethylhexadecylammonium stearate has a specific effect on the mucosal cells of the villi in the pyloric, duodenal and jejunal regions of the gastro-intestinal tract, since the effect is obtained no matter what route is employed for administration of the drug. Large doses of this substance bring about the total destruction of mucosal epithelial cells and lead to the degeneration and collapse of the whole villus, whereas smaller doses appear to reduce the functional efficiency of the mucosal cell probably both in its absorbing as well as its digestive roles. The hypertrophy and hyperplasia following continued oral administration of 0·05 per cent of trimethylhexadecylammonium stearate in the diet seem to represent an effort on the part of the intestine to overcome the functional difficulty imposed by the drug. It would seem akin to the compensatory hypertrophy of a strained myocardium. The thickening is absent in the ileum and colon. This hypertrophy has also been noted occasionally after repeated intraperitoneal injection of the drug. (Intestinal hypertrophy was not produced by new batches of trimethylhexadecylammonium stearate, so that the exact composition of the batch and percentage of impurities may play an important part.)

A substance with such specific pharmacological activity opens up two important research fields. As a research tool it should prove a great aid in the study of intestinal absorption and the many enzymes



Figs. 3–5. Control, medium effect, severe effect (×60)



Figs. 6–8. Control, medium effect, severe effect (×420)

Figs. 4 5, 7 and 8. Jejunums of mice showing moderate and severe damage to villi after subcutaneous injection of trimethy lhexadecylammonium stearate (1·0 gm./kgm.

that are probably concerned in this function. Furthermore, drugs which reduce food absorption may prove valuable in the control of obesity, atherosclerosis and coronary thrombosis.

Finally, the drug was tried on a healthy adult male volunteer. Three consecutive courses of the drug were administered at monthly intervals for periods of three, two and two weeks, respectively, and have resulted in loss of weight of 15, 9 and 7 lb., respectively. The drug was taken in the form of capsules in divided doses, and the average daily doses in the three courses were 1·0, 0·75 and 0·7 gm. This moderate dosage resulted in increase in the bulk and softness of the fæces, whereas a daily dose of 1·5 gm. was associated with mild nausea and fluid stools, but no colic or tenesmus. The effects of the drug were again evidently reversible, since, with over-eating, 7 and 4 lb. weight were put on rapidly at the end of the first and second courses. The total loss in weight was 20 lb.

α,ε-DIAMINOPIMELIC ACID: ITS DISTRIBUTION, SYNTHESIS AND METABOLISM

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THE introduction of paper partition chromatography by Consden *et al.*[1] made it possible to identify the components of many complex organic substances, and introduction of this technique stimulated numerous studies designed to characterize the amino-acid content of many natural products. One such study, first reported by Elizabeth Work[2], indicated the presence of a new ninhydrin positive material, designated as spot No. 17, in hydrolysates of both insoluble cell residues and, although in much lower concentration, in ethanol extracts of *Corynebacterium diphtheriae*. At about the same time an amino-acid with similar chromatographic behaviour was found in hydrolysates of soil[3], rumen contents of sheep (see footnote in Work[4]) and in a lipopolysaccharide fraction from *Mycobacterium tuberculosis*[5]. It remained for Dr. Work, however, in an excellent piece of work, to isolate, characterize and identify this new amino-acid as α,ε-diaminopimelic acid[6].

Properties and Distribution

Properties. Diaminopimelic acid is a neutral compound, moderately soluble in cold water and aqueous alcohol. It is soluble in dilute acid or alkali and is insoluble in ethanol and all organic solvents[4,6].

α,ε-Diaminopimelic acid can be synthesized chemically by the method of Sørenson and Anderson[7] or Sheehan and Bolhofer[8], yielding, in either case, three isomers, the DD, LL and *meso*. Separation of the three isomeric components has been achieved[9] by preparing the diaminopimelic acid diamide dihydrochloride, removing the *meso* diamide by fractional crystallization and resolving the DL racemate with hog kidney amidase[10]. The enzyme-treated racemate was then fractionated on the cation-exchange resin, 'Amberlite *XE*-64', and the respective isomers crystallized. The original procedure of Work *et al.*[9] has been modified by Wade *et al.*[11], who prepared the dicarbobenzoxy derivative which permits separation of the soluble dicarbobenzoxy-*meso*-diaminopimelic acid from the insoluble dicarbobenzoxy-DL-diaminopimelic acid. The diamide was prepared from the above compound, the carbobenzoxy groups removed by hydrogenation and the DL-diaminopimelic acid diamide resolved as described by Work *et al.*[9]. The dissociation constants of *meso* and LL-α,ε-diaminopimelic acid were identical: pk'_1 1·8, pk'_2 2·2, pk'_3 8·8, pk'_4 9·9, but differences in the infra-red spectrum of the two compounds in the region of 6–8μ were observed[11].

The distribution of the various stereoisomers of α-ε-diaminopimelic acid has been studied[12–14], and it is clear that all three isomers are found in micro-organisms. The *meso* isomer or combinations of the *meso* and L-isomers seemed to be the most prevalent form of α,ε-diaminopimelic acid, although some of the *Propionibacteria*, *Streptomyces* and *Clostridia* contain only the L-isomer. The Micromonospora appear to contain all three isomers.

Distribution. Work[6] concluded that α,ε-diaminopimelic acid was probably restricted to protein of bacterial origin, since it had not been reported in the literature concerned with the chromatography of mammalian cell protein hydrolysates or tissue fluids. To check this hypothesis, Work and Dewey[15] conducted a survey of 118 micro-organisms, etc., including fungi, algae, protozoa and viruses, which revealed that α,ε-diaminopimelic acid was present in all the bacteria examined with the exception of the Gram-positive cocci. It was also found in two species of Myxobacteriales and in three species of Myxophyceae. It was absent, however, from yeast, fungi, two strains of protozoa, tobacco mosaic and turnip yellow mosaic virus. Knight[16], in a survey of the amino-acid composition of tomato bushy stunt, influenza *A* and *B*, Shope papilloma, cucumber virus and T_3 bacteriophage did not report the presence of α,ε-diaminopimelic acid in these viruses.

In the original survey[15], α,ε-diaminopimelic acid was reported to be absent from actinomyces and streptomyces but that these organisms contained instead another new amino-acid. This substance was designated as compound *A*. Work[17,18] presented evidence suggesting that compound *A* was a methyl homologue of α,ε-diaminopimelic acid, but subsequent studies[14], using the chromatography system of Rhuland *et al.*[19], revealed that compound *A* was LL-diaminopimelic acid. Cummins and Harris[20,21] and Cummins[22] reported that when α,ε-diaminopimelic acid was found in whole-cell hydrolysates, it was always present in the cell wall.

α,ε-Diaminopimelic acid has been found in soil hydrolysates[3], in caseous tuberculous nodules and caplan nodules[23], in pine tree pollen[24] and in alcohol extracts of neoplastic tissues[25]; but in all cases except the latter the presence of bacterial contamination could not be eliminated. Seibert *et al.*[25], using an α,ε-diaminopimelic acid auxotroph, obtained growth of that organism in the absence of α,ε-diaminopimelic acid when drops of tissue hydrolysates were added to the bioautographic plates. These results must be re-examined with great care, however, since lanthionine was also present in the tissues, and this compound in the presence of trace amounts of α,ε-diaminopimelic acid, for example, when using an unwashed inoculum of *E. coli* 173–25, will support the growth of *E. coli* 173–25 (ref. 26). Seibert *et al.*[25] reported that lanthionine will support the growth of the *E. coli* auxotroph, but this was not confirmed by Rhuland[26] or Meadow *et al.*[27], using the same test organism.

With the finding of Salton[28] that α,ε-diaminopimelic acid exists in the cell walls of *E. coli*, *Salmonella pullorum* and *B. subtilis*, numerous reports have appeared revealing the presence of this amino-acid in various cell fragments. Smith *et al.*[29] found α,ε-diaminopimelic acid in the peptide fraction of a cell wall polysaccharide from *B. anthracis*, and its presence in a soluble peptide, produced by germinating spores of *B. megaterium*, was shown by Strange and Powell[30]. The recent review by Strange[31] should be consulted for a more detailed account of the occurrence of α,ε-diaminopimelic acid in various cell wall peptides of the bacilli. Koch and Dreyer[32] demonstrated the same amino-acid in fragments released by bacteriophage, as have Weidel and Primosigh[33]. A recent publication by Strominger[34] indicated that *meso*-α,ε-diaminopimelic acid occurs in a 'Park's type nucleotide' (Park[35]) in *E. coli*.

With the exception of the demonstration of α,ε-diaminopimelic acid in *Corella ellipsoidia* by Fujiwara and Akabori[36], it appears that the distribution of this amino-acid is limited to the Schizomycetes and the Myxophophyceae, thus lending support to the thesis of Stanier and VanNiel[37] that these groups should be combined into one kingdom, the Monera.

Isolation. The lysine auxotroph, *E. coli* 26–26, isolated by Davis[38], accumulates large amounts of α,ε-diaminopimelic acid in the culture medium and can be used as a source of *meso* and LL-α,ε-diaminopimelic acid. Wright and Cresson[39] used an ion-exchange chromatography procedure involving adsorption of the amino-acid on 'Dowex-1' (OH$^-$) followed by elution with 0·2 *N* hydrochloric acid, removal of anions in the α,ε-diaminopimelic acid fractions with 'Dowex-1' (HCO$_3^-$) and finally deionization with *IR*-4*B* (OH$^-$) and *IRC*-50 (H$^+$). Crystallization of α,ε-diaminopimelic acid was achieved by adding ethanol to concentrated solutions of α,ε-diaminopimelic acid after final de-ionization.

Work and Denman[40] used a somewhat simpler procedure employing acid-washed 'Zeokarb-225'. Contaminating amino-acids, aspartate, glutamate, glycine, alanine and valine were removed first with 1·5 *N* hydrochloric acid. Some 2·5 *N* hydrochloric acid was then added to elute the α,ε-diaminopimelic acid. The amino-acid hydrochloride was crystallized directly from the concentrate or obtained as the free base after removal of chloride ion with deacidite. I have used this same procedure, except that 'Dowex-50' (200–400 mesh) was used as the resin, to obtain α,ε-diaminopimelic acid labelled with carbon-14 from culture filtrates of *E. coli* 26–26 grown on uniformly labelled glucose. Ikawa and O'Barr[41] used the 'Dowex-50' procedure to obtain *meso*-diaminopimelic acid from acid hydrolysates of *Lactobacillus arabinosus*.

Since *E. coli* produces *meso* and LL-α,ε-diaminopimelic acid, both isomers are obtained by these resin procedures, although, as shown by Wright and Cresson[39], the LL isomer may be removed by recrystallization. Hoare and Work[18] attempted to separate the two isomers by the procedure of Work and Denman[40], but were unable to achieve a practical method of separation.

Figueroa and Seibert[42] studied the chromatography of α,ε-diaminopimelic acid on starch columns, and found that this procedure does not separate α,ε-diaminopimelic acid from lysine.

E. coli 26–26 and high-producing mutants derived from it have been used in the commercial production of lysine[43]. In this patented process toluene-treated cells of *A. aerogenes* are added to fermentation broths containing large quantities of α,ε-diaminopimelic acid; the lysine, obtained after decarboxylation of the α,ε-diaminopimelic acid, is isolated by resin chromatography.

Metabolism

Oxidation. Natural diaminopimelic acid from *Corynebacterium diphtheriae*, the *meso* isomer, was shown by Burton[44] to be oxidized by the L-amino-acid oxidase of *Neurospora crassa* with the consumption of one atom of oxygen and the liberation of one molecule of ammonia per molecule of amino-acid (see also Work[6]). Bonetti (personal communication to Work[6]) reported that this same preparation was not oxidized by kidney D-amino-acid oxidase or ophio L-amino-acid oxidase. Subsequent studies by Work[45] revealed that the *Neurospora* oxidase attacks *meso*, LL-α,ε-diaminopimelic acid and the L-diaminopimelic acid-D-monoamide at about the same rate. The L-amino-acid oxidases of viper and rattlesnake attacks the *meso* and L-isomers at a much slower rate than the *Neurospora* oxidase, but metabolized the L-diaminopimelic acid-D-monoamide at a rate almost comparable with the *Neurospora* preparation. The DD-isomer was not attacked nor did it have any effect on the course of oxidation of the other isomers.

Racemization. A racemase, present in acetone-dried cells of *E. coli* 26–26 (lysine auxotroph), converts L-diaminopimelic acid to the *meso* isomer[46]. Subsequent studies[47,48] have revealed some of the properties as well as the distribution of this enzyme. Diaminopimelic acid racemase acts on either *meso* or L-diaminopimelic acid, but has no activity on the DD-isomer. As Antia *et al.*[48] pointed out, the substrate for this enzyme is unusual in that it has two asymmetric carbon atoms which are symmetrical about the γ-carbon atom. The experimental evidence presented indicates that one end of the α,ε-diaminopimelic acid molecule must be in the L-configuration to ensure enzymatic activity.

Diaminopimelic acid racemase was found in almost all bacteria containing α,ε-diaminopimelic acid, although there was no correlation between the type of isomer and racemase content[48]. It is noticeably absent from certain *Clostridia*, but present in several of the Micrococci which do not contain α,ε-diaminopimelic acid.

Decarboxylation. The structural relationship of α,ε-diaminopimelic acid and lysine and the established presence of amino-acid decarboxylases in bacteria[49] suggested an important metabolic relationship between these two amino-acids involving a possible mechanism of lysine biosynthesis in bacteria.

Dewey and Work[50] demonstrated that α,ε-diaminopimelic acid was indeed decarboxylated to lysine by some strains of *E. coli*. The enzyme, as most decarboxylases, was most active in cells grown at acid *p*H, but unlike the previously reported cases, was constitutive rather than adaptive. Davis[38] implicated α,ε-diaminopimelic acid as a precursor of lysine with his finding that a number of lysine auxotrophs accumulated large amounts of α,ε-diaminopimelic acid in the culture supernatant. Vogel[51], studying the metabolism of [4-^{14}C] aspartate in strains of *Hydrogenomonas facilis*, *Pseudomonas fluorescens*, *Azotobacter agilis*, *Agrobacterium radiobacter*, *Alcaligenes faecalis*, *Escherichia coli*, *Micrococcus lysodeikticus*, *Arthrobacter globiformis*, *Bacillus subtilis* and *Streptomyces griseus*, found what appear to be a single over-all mode of lysine biosynthesis, presumably via α,ε-diaminopimelic acid. Similar studies with *Chorella vulgaris* and *Euglena gracilis*[52] revealed that the *Chorella* containing α,ε-diaminopimelic acid synthesize lysine via the α,ε-diaminopimelic acid pathway, while the *Euglena* not containing α,ε-diaminopimelic acid utilized the fungi pathway of lysine biosynthesis, thus lending support to the thesis advanced by Dewey and Work[50] and Davis[38]. Recent experiments by Meadow and Work[53], using an auxotroph of *E. coli* that requires only α,ε-diaminopimelic acid for growth, report that, in this organism, lysine can be synthesized by a metabolic sequence that does not proceed via α,ε-diaminopimelic acid. This conclusion is based on the finding that this auxotroph, *E. coli* 173-25-*D*, grown on glucose labelled with carbon-14, incorporated radioactivity in the cellular lysine, but not the α,ε-diaminopimelic acid.

With the implication of α,ε-diaminopimelic acid as an intermediate in the biosynthesis of lysine it was important not only to determine the distribution of this enzyme, but also to ascertain with certainty that the decarboxylation of α,ε-diaminopimelic acid was not due to a non-specific lysine decarboxylase. That a specific α,ε-diaminopimelic acid decarboxylase was present was clearly demonstrated by Dewey and Work[50] using the lysine auxotroph isolated by Davis[38] which contained lysine decarboxylase, but no α,ε-diaminopimelic acid decarboxylase. Additional proof was obtained by growing *E. coli* in a glucose salts medium which, under these conditions, contains no lysine decarboxylase but does contain the constitutive α,ε-diaminopimelic acid decarboxylase.

Dewey and Work[54] reported the presence of α,ε-diaminopimelic acid decarboxylase in *Aerobacter aerogenes* and, in a later paper, Dewey[55] surveyed a number of strains of the *coli-aerogenes* group and other bacteria for the presence of this enzyme. The results of the survey indicated that the enzyme is widely distributed in the *coli-aerogenes* group. The enzyme was also found in a strain of *Pseudomonas aeruginosa*, but not *Proteus vulgaris*, *Streptococcus faecalis* or *B. subtilis*. Work[56] and Dewey *et al.*[57], in further studies of this type, reported the presence of the enzyme in *Actinomyces* sp., *Rhodopseudomonas spheroides*, *Rhodospirillum rubrum* and in three organisms which do not contain α,ε-diaminopimelic acid, *Sarcina lutea*, *Staphylococcus aureus* and *Streptococcus faecalis*. Powell[58] has reported the enzyme to be present in spores of *B. sphaericus*, as has Willett[59] in *Mycobacterium tuberculosis*.

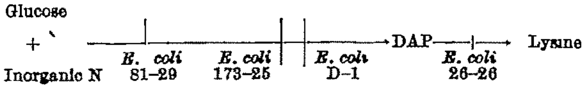
Diaminopimelic acid decarboxylase, unlike most amino-acid decarboxylases, is a constitutive rather than adaptive enzyme[60] and, although formed in cells grown at an acid *p*H, must be assayed at *p*H 6·8 for maximum activity[60]. The enzyme is specific for α,ε-diaminopimelic acid and will not decarboxylate a number of closely relative compounds such as diaminoadipic acid, diaminosuberic acid, α,ε-diamino-β-hydroxypimelic acid, lanthionine or pyridine-2,6-dicarboxylic acid[57,60].

Diaminopimelic acid decarboxylase can be partially purified by fractional extraction with acetone[60] and, although inhibitor studies have shown that it is a very sensitive sulphydryl enzyme, the co-factor requirements have not been ascertained. Pyridoxal phosphate is required for maximal activity[60], but an absolute requirement for this coenzyme has not been demonstrated. The α,ε-diaminopimelic acid and lysine decarboxylase levels in pyridoxin-deficient *E. coli* have been determined[61], but the results failed to show the expected absolute requirement for pyxidoxal phosphate. Similar results were obtained by Powell and Strange[62] with *B. sphaericus*. Meadow and Work[63] studied the α,ε-diaminopimelic acid decarboxylase from *B. sphaericus* in more detail, using a number of pyridoxine inhibitors, but again were unable to demonstrate the absolute requirement of pyridoxal phosphate in this reaction. These workers concluded that there was a low affinity between enzyme and pyridoxal phosphate in *B. sphaericus*.

Initial studies with α,ε-diaminopimelic acid decarboxylase revealed that the enzyme was relatively specific for *meso*-α,ε-diaminopimelic acid, but that acetone-dried cells of *A. aerogenes* decarboxylated LL-α,ε-diaminopimelic acid[18]. Activity against the L-isomer was not always obtained, and Hoare[44] resolved this difficulty with the finding that the apparent decarboxylation of LL-α,ε-diaminopimelic acid was due to its conversion to the *meso* isomer by racemase activity. The D-isomer was not metabolized by α,ε-diaminopimelic acid decarboxylase[46,48].

Transamination. With the widespread distribution of transaminases in bacteria, it is perhaps not unreasonable to expect that α,ε-diaminopimelic acid could act as an amino donor in such a reaction. Evidence demonstrating this reaction was reported by Meadow and Work[64] using acetone-dried cells of *E. coli*, *B. cereus*, *B. sphaericus*, *S. lutea*, *A. aerogenes* and *S. saprophyticus*. Contrary to the racemase, oxidase and decarboxylase reactions, the D-isomer was shown to participate in the transaminase reaction and in the *S. lutea* preparation was transaminated faster than the *meso* isomer. In general, pyruvate, oxalacetate and oxoglutarate act as amino acceptors, but the rate varies with the organism and with the stereoisomer used. Using *B. sphaericus*, Meadow and Work[64] reported that concentrations of pyridoxal phosphate from 1μ to 100μ moles progressively increased the rate of transamination.

Biosynthesis. It is perhaps pertinent to start a discussion of α,ε-diaminopimelic acid biosynthesis with a description of the bacterial mutants which have been used to elucidate in part the mechanism of α,ε-diaminopimelic acid biosynthesis. A number of these mutants have been reported by Davis[38] and enable one to write the following sequence:



E. coli 81–29 requires DAP or lysine if aspartic acid is present in medium.

E. coli 173–25 requires DAP.

E. coli D–1 accumulates *N*-succinyldiaminopimelic acid.

E. coli 26–26 requires lysine, accumulates DAP.

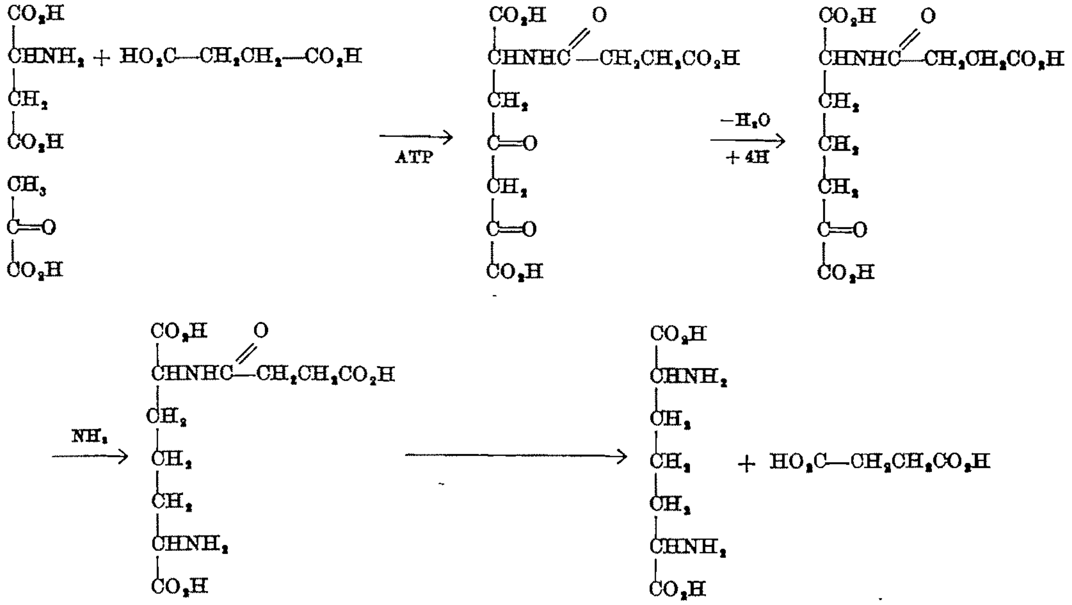
DAP, α,ε-diaminopimelic acid.

As a result of studies of amino-acid biosynthesis with various compounds containing carbon-14, Abelson *et al.*[65] were able to demonstrate that four of the carbon atoms of α,ε-diaminopimelic acid and lysine were derived from aspartic acid. In these studies aliquots of cell hydrolysates were subjected to paper chromatography and the specific activity of the radioactive amino-acids determined directly on the paper. Rhuland and Bannister[66], using *E. coli* 26-26 (accumulates α,ε-diaminopimelic acid) culture filtrates, isolated a compound capable of supporting the growth of *E. coli* 81-29 in the absence of α,ε-diaminopimelic acid. The compound was identified as succinic acid and, as shown by the same authors, was required for maximal synthesis of α,ε-diaminopimelic acid by cell-free preparations of *E. coli* 26-26. In addition to succinic acid, aspartate, pyruvate, adenosine triphosphate and triphosphopyridine nucleotide were also shown to be required in this synthesis. Similar results were reported by Gilvarg[67] with the exception that both di- and tri-phosphopyridine nucleotide were indicated as stimulating α,ε-diaminopimelic acid synthesis, and glutamate instead of succinate was listed as one of the components of the reaction. Subsequent studies by Gilvarg[68] reported the isolation of *N*-succinyl-L-diaminopimelic acid from *E. coli D*-1. This compound was enzymatically hydrolysed to succinic acid and α,ε-diaminopimelic acid by extracts of *E. coli* 26-26 and *E. coli W* (wild type), but not *E. coli D*-1, thus indicating its role as an intermediate in the synthesis of α,ε-diaminopimelic acid and offering an explanation for the function of succinic acid as a growth factor for an α,ε-diaminopimelic acid auxotroph[66]. Additional studies[69] have revealed that the succinyl side-chain is attached to the aspartate portion of N-succinyl-L-diaminopimelic acid.

Gilvarg[70], in a more detailed report of his original observation, confirmed the results of Abelson *et al.*[65], demonstrating that in cell-free extracts, as well as in growing cells, aspartate contributes four of the carbon atoms in the α,ε-diaminopimelic acid molecule. In this study, contrary to the previous report[67], triphosphopyridine nucleotide was indicated as the coenzyme of choice, thus confirming the results of Rhuland and Bannister[66] and Rhuland and Soda[71]. Although the original preliminary report of Gilvarg[67] did not point out which coenzyme was most effective, the coenzyme requirement may depend on the condition of the cells used for preparing α,ε-diaminopimelic acid synthesizing enzymes since Rhuland and Soda[71] showed that diphosphopyridine nucleotide is the preferred coenzyme in extracts from aged cells, but not in extracts from fresh cells. It does not seem likely, however, in view of the results of Rhuland and Soda[71] indicating that glutamate is no more active, with the exception of succinate, than the other Krebs cycle compounds in stimulating α,ε-diaminopimelic acid synthesis, that this compound is used *per se* in this synthesis. It would seem that this evidence and a consideration of the facts that acetyl or formyl aspartate are not used in the reaction[70] along with the isolation of N-succinyldiaminopimelic acid are adequate indications of the role of succinic acid in the reaction sequence under discussion.

The accumulation of 2,N-succinyl-6-ketopimelic acid, in addition to N-succinyldiaminopimelic acid, by *E. coli D*-1 is described by Gilvarg[72]. This compound can be transaminated to N-succinyl-L-diaminopimelic acid by extracts of *E. coli* using glutamic acid as the amino donor. Another brief report[73] described the purification of the transaminase and its specificity for glutamic acid as the amino donor. This transaminase has also been detected in *A. aerogenes*, *A. agile*, *M. lysodeikticus* and *R. spheroides*.

Rhuland and Soda[71] presented evidence revealing that an N-substituted-keto-pimelic acid was made by extracts capable of synthesizing α,ε-diaminopimelic acid. The compound was converted to α,ε-diaminopimelic acid after reductive hydrogenolysis of the 2,4-dinitrophenylhydrazone followed by acid hydrolysis. These results have enabled the above authors to write a tentative mechanism for α,ε-diaminopimelic acid biosynthesis which up to the present time appears to be compatible with the experimental results reported in the literature. The mechanism is as follows.



It is clear from a consideration of this mechanism that the synthesis of α,ε-diaminopimelic acid from small carbon fragments involves seven or eight enzymatic steps with the formation of some new and novel organic compounds. However, it is interesting to note that the enzymatic studies reported thus far have not indicated co-factor requirements other than triphosphopyridine nucleotide. I have achieved a ten-fold purification, involving dialysis, protamine and ammonium sulphate fractionation, of the α,ε-diaminopimelic acid synthesizing complex without demonstrating the need for additional co-factors. The abstract of Peterkofsky and Gilvarg[73] indicated a 150-fold purification of the N-succinyl-ε-ketopimelic-glutamic transaminase; but did not mention a pyridoxal phosphate requirement for this system.

Experiments similar to those described by Abelson *et al.*[65] have been interpreted by Meadow and Work[74] to indicate two pathways of α,ε-diaminopimelic acid biosynthesis. These conclusions were based on the fact that *E. coli* 26-26 grown with unlabelled glucose and aspartate labelled with carbon-14 and/or pyruvate incorporated radioactivity into cellular α,ε-diaminopimelic acid but not in the exogenous α,ε-diaminopimelic acid. Use of trace amounts of labelled acetate under the same conditions, however, resulted in radioactivity in both the cellular and extracellular α,ε-diaminopimelic acid. These observations and the fact that extracellular α,ε-diaminopimelic acid was not produced until the period of maximum growth was attained enabled the above authors to conclude that cellular α,ε-diaminopimelic acid was synthesized by one metabolic pathway and that extracellular α,ε-diaminopimelic acid was synthesized from different precursors.

These studies do not negative any of the findings on α,ε-diaminopimelic acid biosynthesis described above; but it would appear that further investigations will be necessary to establish with certainty that two different synthetic pathways for α,ε-diaminopimelic acid are operative.

Elucidation of the steps involving activation of succinate, aspartate and pyruvate and the identification of all the components in this metabolic sequence, as well as the many aspects of α,ε-diaminopimelic acid metabolism still to be investigated; activation and incorporation into peptides and bacterial cell walls, its role in lipopolysaccharide complexes and the specific function of each isomer, will provide a very interesting field of study.

Added in proof. Gilvarg (*J. Biol. Chem.*, 234, 2955; 1959) has reported a more detailed account of the production and isolation, as well as the chemical synthesis, of N-succinyl-L-diaminopimelic acid. Contrary to his previous report[68], he indicates that N-succinyl-L-diaminopimelic acid is actually produced by mutant *E. coli* D-1$_{(\alpha)}$. Rhuland and Hamilton (*Biochim. Biophys. Acta*, 36, 559; 1959) report that *E. coli* 173-25, the α,ε-diaminopimelic acid auxotroph, contains all the enzymes necessary to synthesize α,ε-diaminopimelic acid. In addition to the synthesis of LL-diaminopimelic acid by this organism, another substance, presumably N-succinyl-diaminopimelic acid, is produced.

[1] Consden, R., Gordon, A. H., and Martin, A. J. P., *Biochem. J.*, 38, 224 (1944).

[2] Work, E., *Biochimica et Biophysica Acta*, 3, 400 (1949).

[3] Bremmer, J. M., *Biochem. J.*, 47, 538 (1950).

[4] Work, E., *Nature*, 165, 74 (1950).

[5] Asselineau, J., Choucroun, N., and Lederer, E., *Biochimica et Biophysica Acta*, 5, 197 (1950).

[6] Work, E., *Biochem. J.*, 49, 17 (1951).

[7] Serenson, S. P. L., and Anderson, A. C., *Hoppe-Seyl. Z.*, 56, 250 (1908).

[8] Sheehan, J. C., and Bolhofer, W. A., *J. Amer. Chem. Soc.*, 72, 2786 (1950).

[9] Work, E., Birnbaum, S. M., Winitz, M., and Greenstein, J. P., *J. Amer. Chem. Soc.*, 77, 1916 (1955).

[10] Hamer, D., and Greenstein, J. P., *J. Biol. Chem.*, 193, 81 (1951).

[11] Wade, R., Birnbaum, S. M., Winitz, M., Koegel, R. J., and Greenstein, J. P., *J. Amer. Chem. Soc.*, 79, 648 (1957).

[12] Hoare, D. S., and Work, E., *Biochem. J.*, 60, ii (1955).

[13] Hoare, D. S., and Work, E., *Biochem. J.*, 61, 562 (1955).

[14] Hoare, D. S., and Work, E., *Biochem. J.*, 65, 441 (1957).

[15] Work, E., and Dewey, D. L., *J. Gen. Microbiol.*, 9, 394 (1953).

[16] Knight, C. A., "Adv. in Virus Res.", 2, 153 (1954).

[17] Work, E., *J. Gen. Microbiol.*, 9, ii (1953).

[18] Work, E., Proc. 6th Int. Microbiol. Congr. (Rome), 1, 161 (1953).

[19] Rhuland, L. E., Work, E., Denman, R. F., and Hoare, D. S., *J. Amer. Chem. Soc.*, 77, 4844 (1955).

[20] Cummins, C. S., and Harris, H., *J. Gen. Microbiol.*, 13, iii (1955).

[21] Cummins, C. S., and Harris, H., *J. Gen. Microbiol.*, 14, 583 (1956).

[22] Cummins, C. S., *Int. Rev. Cytology*, 5, 25 (1956).

[23] Consden, R., and Glynn, L. E., *Lancet*, 268, 943 (1955).

[24] Cummings, M. M., and Hudgins, P. C., Abst. Amer. Soc. Clin. Invest. (1958).

[25] Seibert, F. B., Soto-Figueroa, E., Miller, E. E., and Seibert, M. V., *Growth*, 18, 145 (1954).

[26] Rhuland, L. E., *J. Bact.*, 73, 778 (1957).

[27] Meadow, P., Hoare, D. S., and Work, E., *Biochem. J.*, 66, 270 (1957).

[28] Salton, M. R., *Biochimica et Biophysica Acta*, 10, 512 (1953).

[29] Smith, H., Strange, R. E., and Zwartouw, H. T., *Nature*, 178, 865 (1956).

[30] Strange, R. E., and Powell, J. F., *Biochem. J.*, 58, 80 (1954).

[31] Strange, R. E., *Bact. Rev.*, 23, 1 (1959).

[32] Koch, G., and Dreyer, W. J., *Virol.*, 6, 291 (1958).

[33] Weidel, V. W., and Primosigh, J., *Z. Naturforschung*, 12b, 419 (1957)

[34] Strominger, J. L., Scott, S. S., and Threnn, R. H., *Fed. Proc.*, 18, 334 (1959).

[35] Park, J. W., *J. Biol. Chem.*, 194, 877 (1951).

[36] Fujiwara, T., and Akabori, S., *J. Chem. Soc.* (*Japan*), 75, 990 (1954). *Chem. Abst.*, 49, 3325h (1955).

[37] Stanier, R. Y., and VanNiel, C. B., *J. Bact.*, 42, 437 (1941).

[38] Davis, B. D., *Nature*, 169, 534 (1952).

[39] Wright, L. C., and Cresson, E. L., *Proc. Soc. Exp. Biol. and Med.*, 82, 354 (1953).

[40] Work, E., and Denman, R. F., *Biochimica et Biophysica Acta*, 10, 183 (1953).

[41] Ikawa, M., and O'Barr, J. S., *J. Biol. Chem.* 213, 877 (1955).

[42] Figueroa, E. S., and Seibert, F. B., *Proc. Soc. Exp. Biol. and Med.*, 83, 535 (1953).

[43] Casida, L. E., U.S. Patent Office 2,771,396 (Nov. 20, 1956).

[44] Burton, K., *Biochem. J.*, 50, 258 (1951).

[45] Work, E., *Biochimica et Biophysica Acta*, 17, 410 (1955).

[46] Hoare, D. S., *Biochem. J.*, 59, xxii (1955).

[47] Hoare, D. S., and Work, E., *J. Gen. Microbiol.*, 15, xiii (1956).

[48] Antia, M., Hoare, D. S., and Work, E., *Biochem. J.*, 65, 448 (1957).

[49] Gale, E. F., "Adv. in Enzymol.", 6, 1 (1946).

[50] Dewey, D. L., and Work, E., *Nature*, 169, 533 (1952).

[51] Vogel, H. J., *Fed. Proc.*, 18, 345 (1959).

[52] Vogel, H. J., *Biochimica et Biophysica Acta*, 34, 282 (1959).

[53] Meadow, P., and Work, E., *Biochem. J.*, 72, 400 (1959).

[54] Dewey, D. L., and Work, E., 2nd Int. Congr. Biochem., 77 (1952).

[55] Dewey, D. L., *J. Gen. Microbiol.*, 11, 307 (1954).

[56] Work, E., "Amino Acid Metabolism", ed. by McElroy, W., and Glass, B., 462 (Johns Hopkins Press, 1955).

[57] Dewey, D. L., Hoare, D. S., and Work, E., Proc. Sixth Int. Microbiol. Congr. (Rome), 1, 261 (1953).

[58] Powell, J. F., *Biochem. J.*, 70, 91 (1958).

[59] Willet, H. P., *Proc. Soc. Exp. Biol. and Med.*, 99, 177 (1959).

[60] Dewey, D. L., Hoare, D. S., and Work, E., *Biochem. J.*, 58, 523 (1954).

[61] Denman, R. F., Hoare, D. S., and Work, E., *Biochimica et Biophysica Acta*, 16, 442 (1955).

[62] Powell, J. F., and Strange, R. E., *Biochem. J.*, 65, 700 (1958).

[63] Meadow, P., and Work, E., *Biochemica et Biophysica Acta*, 29, 180 (1958).

[64] Meadow, P., and Work, E., *Biochimica et Biophysica Acta*, 28, 596 (1958).

[65] Abelson, P. H., Bolton, E., Britten, R., Cowie, D. B., and Roberts, R. B., *Proc. U.S. Nat. Acad. Sci.*, 39, 1020 (1953).

[66] Rhuland, L. E., and Bannister, B., *J. Amer. Chem. Soc.*, 78, 3548 (1956).

[67] Gilvarg, C., *Fed. Proc.*, 15, 261 (1956).

[68] Gilvarg, C., *Biochimica et Biophysica Acta*, 24, 216 (1957).

[69] Gilvarg, C., Edelman, J., and Kindler, S. H., Fourth Int. Congr. Biochem. Sept., 10 (1958).

[70] Gilvarg, C., *J. Biol. Chem.*, 233, 1501 (1958).

[71] Rhuland, L. E., and Soda, J. A., *J. Bact.*, 78, 400 (1959).

[72] Gilvarg, C., *Fed. Proc.*, 17, 229 (1958).

[73] Peterkofsky, B., and Gilvarg, C., *Fed. Proc.*, 18, 301 (1959).

[74] Meadow, P., and Work, E., *Biochem. J.*, 72, 396 (1959).

LETTERS TO THE EDITORS

ASTROPHYSICS

Low-Energy X-Rays from Interplanetary Space

It is generally believed that the Sun is a source of highly ionized gas which streams outward through the solar system at all times and in all directions[1]. At the orbit of the Earth, even under the quietest solar conditions, the velocity and density appear to be of the order of 500 km./sec. and 10^2 to 10^3 particles/cm.^{3} respectively. (When the Sun is very active velocities greater than 1,500 km./sec. and densities up to 10^5 cm.^{3} may be produced sporadically.) Theoretically[2], it is expected that this steady gas stream will finally become unstable at some radial distance from the Sun and establish a region of highly disordered interplanetary magnetic field and plasma.

I wish to point out that energy interchange processes between the heavy ions and the electrons of the plasma which may be expected to occur in this disordered region of hydromagnetic turbulence should lead to a diffuse source of low-energy cosmic X-rays which may be observable at altitudes greater than about 120 km. Exchange processes available, in addition to the Fermi and betatron acceleration mechanisms, include excitation of plasma oscillations[3] and a general process described by Veksler[4] which may operate as a plasma encounters any inhomogeneous magnetic field. The Veksler mechanism can, in principle, lead to complete interchange of energy between the protons and the electrons of the streaming solar plasma, producing thereby electrons with energies somewhat in excess of 1 keV. The total energy W_γ in the form of bremsstrahlung photons radiated by these electrons per c.c. of the disordered region, if only $Z = 1$ ions are considered, is given by:

$$W_\gamma = 11 \cdot 9 \times 10^{-20} N^2 \overline{T}^{\frac{1}{2}} \text{ ergs/cm.}^3\text{/sec.}$$

Presence of He and higher Z ions in the interplanetary gas will be neglected although such ions can probably contribute appreciably to the radiated power. $\overline{T}$ is the average energy of the electrons in ergs.

I now consider only photons between T and αT, where α is an instrumental cut-off, taking the electrons to be mono-energetic with energy T. The total number of such photons arriving near the Earth may be computed if we assume spherical symmetry, a disordered region extending radially[2] from 2 to 5 A.U., and complete energy exchange (without the inverse process) over a distance short compared to the thickness of the disordered region. Absorption is negligible since the total Thompson scattering probability in a path 1 A.U. in length is only $\sim 10^{-7}$. Under these conditions, the X-ray number flux F_γ near the orbit of Earth is given by:

$$F_\gamma = \frac{11 \cdot 9 \times 10^{-20} \log_e\left(\frac{1}{\alpha}\right)}{T^{\frac{1}{2}}} \, a^4 \int_{2a}^{5a} N_0^2 \, r^{-4} \, dr$$

where a is the Sun–Earth distance $= 1 \cdot 5 \times 10^{13}$ cm. = 1 A.U. and N_0 is the plasma particle density near the Earth.

If we consider X-rays between 10 and 50 Å., which may be readily measured with rocket or satellite-borne proportional counters, then $\alpha \approx 0 \cdot 2$ and with $N_0 = 10^2$ we find $F_\gamma \simeq 23$ X-rays/cm.^{2}/sec. corresponding to an energy flux of $\sim 3 \times 10^{-8}$ ergs/cm.^{2}/sec. Note that if N_0 is closer to 10^3 than 10^2 the flux will be $\sim 3 \times 10^{-6}$ ergs/cm.^{2}/sec. or $2 \cdot 3 \times 10^3$ photons/cm.^{2}/sec. (Radiation which may originate from individual streams of exceptional velocity and density such as are produced during periods of great solar activity is discussed elsewhere. Reiffel, L. (to be published).) These are, of course, upper limits to the flux, on the assumption that complete energy transfer occurs, which is obviously improbable. However, on the night side of the Earth, provided background radiation from celestial sources is not serious, rather high sensitivity might be achieved, hence the radiation may be detectable even in the event of a low energy interchange efficiency. Solar background radiation, backscattered into the detectors, would be low because of the small total scattering probability per A.U. mentioned earlier. Collimated detectors might be used to advantage to look for structure, for example, higher intensity in the plane of the ecliptic or 'patchiness' in the interplanetary gas X-ray source, and to discriminate against stellar backgrounds and auroral X-rays from the Earth.

To my knowledge, the only experimental information on X-rays in the night sky is the negative result obtained in a measurement briefly mentioned by Friedman[5] and obtained in the course of the N.R.L. rocket programme. The upper limit, established by the lack of a measurable flux, in this case was 10^{-8} ergs/cm.^{2}/sec. per Å. in the region around 50 Å. The counters used for the measurement were presumably of the type described by Byram *et al.*[6] and, as such, were more appropriate to detecting the high fluxes associated with direct exposure to solar X-rays. The upper limit quoted above corresponds to an energy flux, calculated on the basis of a thin target bremsstrahlung spectrum, of about 2×10^{-6} ergs/cm.^{2}/sec. over the wave-length range 10–50 Å. This is just about the upper limit calculated for the X-rays of interplanetary origin on the assumption of the higher gas density. Presumably counters specifically designed for this task could be made a great deal more sensitive since the effective area of the counters used by Byram *et al.* was only of the order of $0 \cdot 05$ cm.^{2} while the efficiency was about 5 per cent.

The possibility of observing radio noise from the mechanism described above will be discussed in a subsequent communication.

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[1] Biermann, L. Z., *Astrophys. J.*, **29**, 274 (1957); *Naturforsch.*, **7**, *a*, 127 (1952); *Observatory*, **77**, 109 (1957).

[2] See especially Parker, E. N., "The Plasma in a Magnetic Field", 80 (Stanford Univ. Press, Stanford, Calif.); *Astrophys. J.*, **128**, 674 (1958).

[3] Parker, E. N., *Phys. Rev.*, **112**, 1429 (1958).

[4] Veksler, V. I., *Soviet Phys.—Doklady*, **3**, 84 (1958).

[5] Friedman, H., *Inst. Rad. Eng.*, **47**, 278 (1959).

[6] Byram, E. T., Chubb, T. A., and Friedman, H., *J. Geophys. Res.*, **61**, 251 (1956).

GEOPHYSICS

Observations on the Solar Eclipse of October 2

During the recent solar eclipse of October 2, 1959, measurements were made at the U.S. Air Force Cambridge Research Center's radio astronomy site at Hamilton, Massachusetts. At this location the Sun rose partially eclipsed and 10 min. later it was totally eclipsed at an elevation of about 1°.

Having tracked the Sun from sunrise for a week prior to the eclipse, average refraction corrections were worked out to permit accurate antenna pointing and tracking of the Sun.

Measurements were made simultaneously at 224 and 1,300 Mc./s., using an 84-ft. paraboloid, and at 3,000 Mc./s. using an 8-ft. dish located 80 ft. east of the 84-ft. antenna.

Information was obtained from the records of the eclipse and in comparison with records of days both before and after the eclipse (control days).

On the eclipse day, the Sun was quiet. In fact, it was at its coldest level for the two weeks observed. However, interferometric solar maps at 3,000 Mc./s. (R. Bracewell, personal communication), at 9,300 Mc./s. (J. F. Denise, personal communication) and at 1,420 Mc./s. (W. N. Christiansen, personal communication) indicate the presence of plage areas on both the east and west limbs of the Sun. From this, one would expect the apparent solar diameter to be larger than the quiet Sun in the shorter wave-length region, with high-intensity levels extending beyond the solar disk.

This accounts for the fact that the residual energy recorded at totality at both 3,000 Mc./s. and 1,300 Mc./s. of 20 per cent of the uneclipsed Sun is greater than the signals recorded during low sunspot cycle year eclipses. At 224 Mc./s., however, with radiation originating in the corona little affected by the plage areas, and with interferometric maps plotted at this frequency-range at Nançay, France, for example, indicating no point source of solar activity, the residual energy-level of about 35 per cent at totality was not unlike the value for low sunspot cycle year eclipse records.

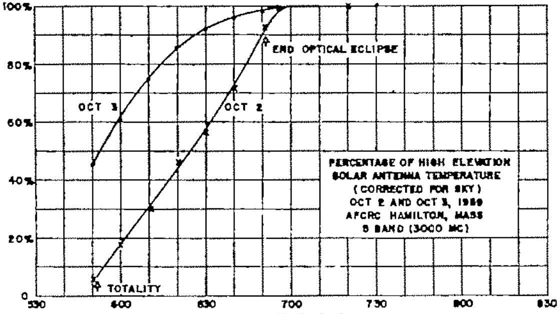

Fig. 1

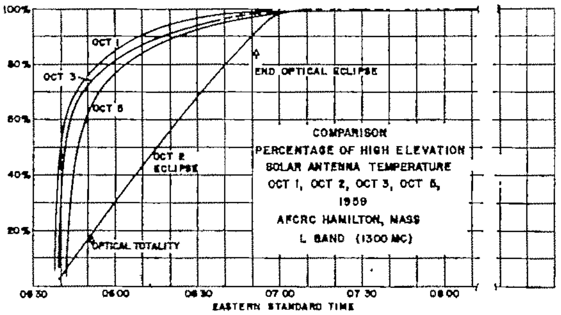

Fig. 2

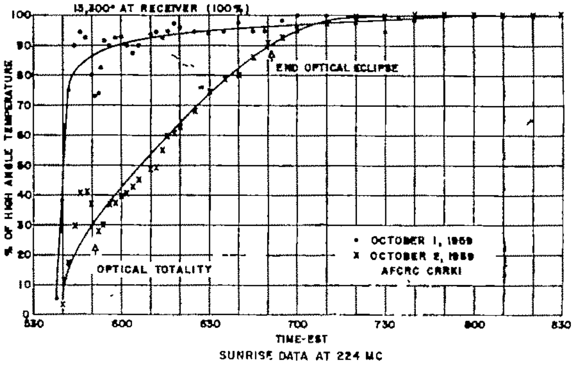

Fig 3

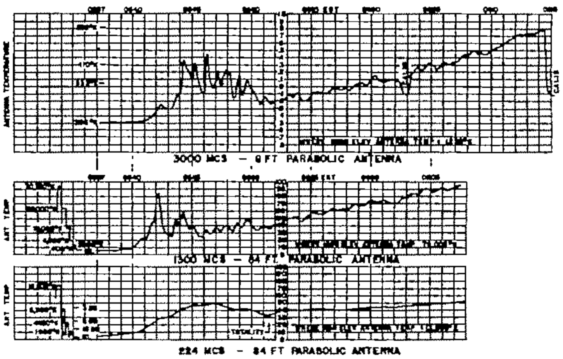

Fig. 4

Energy-levels during the recent eclipse are plotted in Figs. 1, 2 and 3.

Fig. 4 is a record of the signals received in the early part of the eclipse period. Since totality occurred when the Sun's elevation was approximately one degree, low-angle effects were evident. Large scintillations were seen starting at about 0542 E.S.T., especially at the higher frequencies. At this time, the Sun was rising with the east limb as yet uneclipsed. Two radio-frequency active plage areas in this region account for the high-amplitude scintillations. Normally with an uneclipsed solar disk the active plage areas act as point sources and at elevations up to 4 or 5° produce fairly deep fluctuations as, indeed, they did on all days observed. However, none could approach the violence of the 1,300 Mc./s. fluctuation at 0542 E.S.T., Oct. 2, 1959, in terms of percentage of antenna temperature, which was 5 or 6 times the mean curve temperature. Throughout the entire period illustrated, scintillations were high due to early covering and later uncovering of point sources on the Sun with scintillations existing even during 'totality'.

Individual fluctuations at 1,300 Mc./s. and 3,000 Mc./s. were remarkably well correlated in detail, in spite of the separation of the two antennæ and the difference in size of antenna apertures. Thus the peaks and nulls of one frequency are well matched by the other throughout the entire control period.

During the control period, no scintillations were recorded at 224 Mc./s. This is consistent with the quiet nature of the Sun and its large angular diameter at this frequency.

From the scintillation records, two points are evident. The first—the shadow patterns at 1,300 Mc./s. and 3,000 Mc./s. (generated by the radio-frequency active areas on the Sun, which interfering with one another normally decrease the amplitude of the scintillations) are not dependent on frequency. This point is somewhat at variance with work reported by Kazès[1]. The second—within the distance between the centres of the two antennæ, a single shadow pattern exists. This latter observation is generally in accord with work of Steinberg and Kazès[2].

A more detailed analysis of the eclipse and scintillation data will appear at a later date.

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[1] *C.R. Acad. Sci., Paris*, **245**, 636 (1957).

[2] *C.R Acad. Sci., Paris*, **245**, 782 (1957).

Rare Hiss, Earth Currents and Micropulsations on November 27, 1959

ON November 27, 1959, a rare signal was heard on the chorus-whistler equipment[1] at College, Alaska. Beginning as a whistle of perhaps 2 kc./s. at approximately 1351 A.S.T. (2351 G.M.T.) on November 27, 1959, a tone rose slowly to about 10 kc./s., becoming a hiss lasting for what seemed to be several minutes. At the same time magnetic field micropulsations[2] with periods of 10–20 sec. were initiated with a commencement magnitude of about 2·5γ falling to 0·4γ in about 5 min.

An unusually abrupt sudden commencement on the Earth current record[3] at College was also observed at 1351 A.S.T. Although sudden commencements are not rare in the Earth current records, they are usually preceded by some sort of activity. During the hour before the sudden commencement the maximum range of activity in the geographical north–south component was 16 mV./km. At 1351 the trace was abruptly displaced—572 mV./km. In less than 30 sec. the trace recovered and went + 410 mV./km. A negative deflexion of the trace corresponds to a northerly directed electron current in the Earth. For the next 3 hr. the trace oscillated about 150 mV./km. with a dominant period of about 2 min. Strong activity continued through the night, and storms occurred during the next two nights.

A red auroral arc was observed later that day commencing in the north about 1645 A.S.T., reaching the zenith at about 1720 and fading by 1745.

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[1] Pope, J. H., "An Investigation of Whistlers and Chorus at High Latitudes", Sci. Rep. No. 4, *AF* 19(604)-1859, Geophys. Inst., Univ. of Alaska (1959).

[2] Campbell, W. H., *J. Geophys. Res.*, **64**, 1819 (1959).

[3] Hessler, V. P., and Wescott, E. M., "Rapid Fluctuations in Earth Currents at College", Sci. Rep. No. 1, *AF* 19(604)-3075, Univ. of Alaska (Jan. 1959).

Continental Drift

IN recent years the question of continental drift has received renewed attention mainly as a result of palæomagnetic studies. The general consensus of opinion appears now to look with favour upon this hypothesis. No attempt will be made here to summarize all the supporting evidence—rather the purpose of this communication is to point out a difficulty with such a theory. This is based in part on measurements of the flow of heat through the Earth. Certainly far too few heat-flow measurements have been made to draw any quantitative conclusions. Nevertheless, in spite of quite a large range in the measurements of the flow of heat through the ocean floors, the average flow from oceanic and continental areas is approximately the same. There are also many cases where the oceanic flow of heat is greater than the average value through the continents. These results were unexpected on the basis of the radioactive content of typical oceanic and continental crustal rocks existing near the Earth's surface.

It is a very real problem to find sources of heat beneath the ocean floors. The whole of the observed continental heat flow could be produced by the uranium, thorium and potassium contained in about 25 km. of granite. Granitic type rocks are almost completely absent under the oceans—typical oceanic rocks being basalt the radioactive content of which is only about 30 per cent of that of continental rocks. Bullard, Maxwell and Revelle[1] have shown that heat production in the sediments due to biological activity, radioactivity and other processes such as chemical weathering cannot account for more than 1 per cent of the observed values. Again, the oceanic crust, being relatively thin and composed mainly of basaltic materials, can produce at most 10 per cent of the observed values. The remaining heat must therefore come from the mantle. There is thus a striking difference between the mantle under the oceans and under the continents. Hence, if a continent, with its more radioactive crust, were to drift over an oceanic area, with its more radioactive mantle, much higher continental heat flows than are observed would be expected.

The distribution of heat sources in the mantle under the oceans is quite critical if melting at depth is to be avoided. A distribution of radioactivity to depths greater than 200 km. would lead to temperatures above the melting point. On the other hand, a distribution of radioactivity through the upper 200 km. of the mantle would require a much greater radioactive concentration than that found in ultrabasic rocks or stony meteorites. A deeper distribution of radioactive heat sources without producing melting would be possible if rocks at depth have a much higher thermal conductivity than is generally assumed, or by convection in the mantle.

It may be argued that the variations observed in the oceanic heat flow are due to convection currents in the mantle. Thus, for example, the recent high values found by Von Herzen[2] along the East Pacific Rise with areas of low heat flow flanking it may be due to a large convective cell in the upper part of the mantle. But if such an explanation is true, it is difficult to see why convection cells should be confined to the oceanic mantle and not be present under the continents, where the average heat-flow values show considerably less variation.

The concentration of uranium, thorium and potassium in the outer layers of the Earth must have occurred during its very early history (whether a hot or cold origin is postulated), the thickness of this outer layer being a few hundred km. The mantle under the oceans is thus presumably 'primeval', while that under the continents is 'anomalous'. However, the continents were formed, whether initially or whether, as advocated by Wilson[3], for example, they have grown from nuclei throughout geological time drawing their material from the mantle, the fact remains that the constitution of the upper part of the mantle is at present very different beneath the oceans and beneath the continents. It is this fundamental difference that makes it very difficult to see how continental drift can have occurred —unless the continents drag the upper few hundred km. of the mantle along with them, which seems extremely unlikely.

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[1] Bullard, E. C., Maxwell, A. E., and Revelle, R., "Adv. in Geophys.", 3, 153 (1956).

[2] Von Herzen, R., *Nature*, 183, 882 (1959).

[3] Wilson, J. Tuzo, *Amer. Sci.*, 47, 1 (1959).

Isotopic Distribution in a Greenland Iceberg

THE heavy-oxygen content in the ice of the Greenland ice cap is extremely low[1-3]. This is part of a common feature showing decreasing $H_2{}^{18}O$ content in fresh water when going towards a colder climate[1].

At a given location the seasonal variation in temperature causes a similar variation in the heavy-oxygen content of the precipitation[3,4]. On the Greenland ice cap, where the precipitation accumulates, this latter variation has been refound by investigation of stratigraphical series of snow and ice samples from a borehole 400 m. deep[3]. Parallelism between the deuterium and oxygen-18 content has been shown in snow samples from the Greenland ice cap[5]. The present work shows this parallelism to be conserved in the ice during several hundred years.

The samples investigated were collected during the North American Arctic Institute Greenland Expedition 1958 headed by Dr. P. F. Scholander. The deuterium analyses were made at Centre d'Etudes Nucléaires de Saclay by the technique described in ref. 6, while the oxygen-18 measurements were carried out at the University of Copenhagen by the technique described in ref. 7.

The samples were all taken from one piece of ice emitted from the Ingnerit glacier in West Greenland (70·9° N.). This piece was part of a larger amount of ice the average age of which has been determined by Scholander and co-workers by means of the carbon-14 method using the technique described in erf. 8. The piece of ice in question was white with mostly round bubbles. Three parallel layers of clear ice were situated 33 and 25 cm. from each other. The samples were taken along a line perpendicular to these layers.

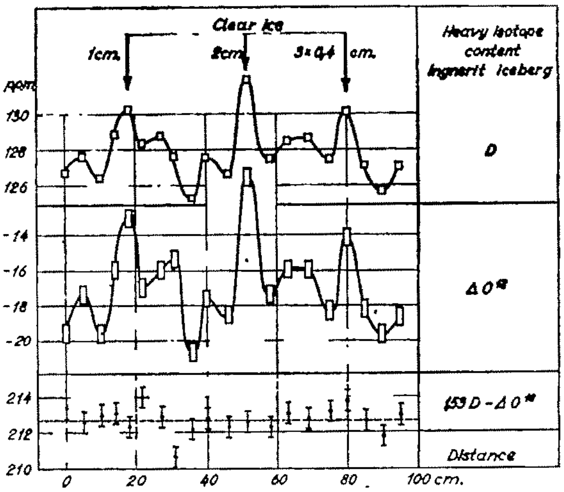

Fig. 1

In Fig. 1 the deuterium content (D) and the deviation of the oxygen-18 content (ΔO^{18}) of the sample from a standard are plotted in units of p.p.m. against the distance of the sample from an arbitrary zero point. In the lower part of Fig. 1 the relation between D and ΔO^{18} is seen to be linear within the measuring accuracy. With the use of our particular standard for oxygen-18 the linear relation is $1 \cdot 53 \text{ D} - \Delta O^{18} = 212 \cdot 7$. This shows complete parallelism between the variations in deuterium and oxygen-18, and, furthermore, conservation of isotopic anomalies in the ice during several hundred years.

The highest heavy isotope contents are found in the parallel layers of clear ice probably originating from summer-melting at the time of formation of the ice. This is due to evaporation from the wet snow; the light isotopic component of the water being the most volatile one.

We wish to thank Dr. P. F. Scholander and Dr. J. Koch for their interest in the work. The project was sponsored by the Arctic Institute of North America with principal support from the U.S. Office of Naval Research. Financial support was also granted by Statens almindelige Videnskabsfond, Copenhagen.

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[1] Dansgaard, W., *Geochim. Cosmochim. Acta*, 6, 241 (1954).

[2] Epstein, S., U.S. Nat. Acad. Sci., Nuclear Science Series, Rep. No. 19 (1956).

[3] Epstein, S., and Sharp, R. P., I.G.Y. Bull., Nat. Acad. Sci., 21 (March 1959).

[4] Dansgaard, W., *Fysisk Tidsskrift*, 56, 49 (1958).

[5] Kulp, J. L., Giletti, B. J., and Erickson, G. P., Lamont Geol. Observatory (Oct. 31, 1957).

[6] Botter, R., and Nief, G., Joint Conference on Mass Spectrometry, September 1958 (Pergamon Press) (in the press).

[7] Dansgaard, W., *Tellus*, 5, 461 (1953).

[8] Coachman, L. K., Hemmingsen, E., Scholander, P. F., Enns, T., and de Vries, H., *Science*, 127, 1288 (1958).

PHYSICS

A Very General Class of Exact Solutions in Concentration-dependent Diffusion

THE mathematics of concentration-dependent diffusion has been found in recent years to be important in problems of diffusion in polymers[1–3] and in the problems of the transport of water in its various phases in soils and other porous materials[4–7]. Almost without exception, it has been necessary to use numerical methods to secure the necessary solutions[2,3,8,9]. According to Crank[10], the only known 'formal solutions' are due to Fujita[11,12]. They provide solutions of the equation:

$$\frac{\partial\theta}{\partial t} = \frac{\partial}{\partial x}\left(D(\theta)\frac{\partial\theta}{\partial x}\right) \qquad (1)$$

subject to the conditions:

$$\begin{aligned} \theta = 0,\ \ x > 0,\ \ t = 0\,, \\ \theta = 1,\ \ x = 0,\ \ t > 0\,; \end{aligned} \qquad (2)$$

and are for the following $D(\theta)$ functions:

$$D = D_0/(1-\lambda\theta);\ D = D_0/(1-\lambda\theta)^2;$$
$$D = D_0/(1 + 2a\theta + b\theta^2).$$

Here we report the existence of a very general class of $D(\theta)$ functions which yield exact solutions of (1) subject to conditions (2). Fujita's $D(\theta)$'s are contained in this class. A similar very general class of $D(\theta)$ functions yields exact solutions of (1) subject to another set of practically important governing conditions:

$$\theta = 0, x > 0 \text{ and } \theta = 1, x < 0, t = 0;$$
$$\int_0^1 x\,d\theta = 0, t \geqslant 0. \qquad (3)$$

The substitution:

$$\varphi = xt^{-1/2}$$

enables (1) subject to (2) or (3) to be reduced to[8]:

$$\int_0^\theta \varphi d\theta = -\ 2D\,d\theta/d\varphi.$$

That is:

$$D = -\ \tfrac{1}{2}\int_0^\theta \varphi\,d\theta\,.\,d\varphi/d\theta.$$

It follows that the solution of (1) subject to (2) or (3), $\varphi(\theta)$, will exist in exact form, so long as $D(\theta)$ is of the form:

$$D = -\ \tfrac{1}{2}\int_0^\theta F\,d\theta\,.\,dF/d\theta, \qquad (4)$$

where F is an analytical (I use this adjective to describe infinitely differentiable functions of real variables) function of θ which satisfies the following conditions: (*a*) the integral $\int_0^\theta F\,d\theta$ exists; (*b*) $dF/d\theta \leq 0$; (*c*) for (1) subject to (2), $F(1) = 0$; (*d*) for (1) subject to (3), $\int_0^1 F\,d\theta = 0$. (Note that D will be expressible in terms of known functions so long as F is integrable in terms of known functions.)

A very large class of F functions satisfies these conditions, so that, accordingly, there exists a very large class of D functions which lead to exact solutions of (1) subject to the appropriate conditions. There is thus an *embarras de richesse* of exact solutions available. A few very simple results are given in Tables 1 and 2.

Table 1. SOME SIMPLE CASES OF EXACT SOLUTIONS OF (1) SUBJECT TO (2)

D	φ	Remarks
$\frac{n}{2}\theta^n\left(1-\frac{\theta^n}{1+n}\right)$	$1-\theta^n$	$n > 0$
$\frac{n}{2}\theta^{-n}\left(\frac{\theta^{-n}}{1-n}-1\right)$	$\theta^{-n}-1$	$0 < n < 1$
$\frac{1}{2}\sin^2\frac{\pi\theta}{2}$	$\cos\frac{\pi\theta}{2}$	
$\frac{1}{2}e^{2-\theta}(1-e^{-\theta}-\theta/e)$	$e^{1-\theta}-1$	

Table 2 SOME SIMPLE CASES OF EXACT SOLUTIONS OF (1) SUBJECT TO (3)

D	φ	Remarks
$\frac{n\,(n+1)}{2}\theta^n\,(1-\theta^n)$	$1-(n+1)\,\theta^n$	$n > 0$
$\frac{n}{2\,(1+n)}[(1-2\theta)^{n-1} - (1-2\theta)^{2n}]$	$(1-2\theta)^n$	$n = 1, 3, 5, 7, ..$
$\frac{n}{2\,(1-n)}\theta^{-n}\,(\theta^{-n}-1)$	$\theta^{-n}-\frac{1}{1-n}$	$0 < n < 1$
$\frac{e^\theta}{2}[(e-1)\theta+1-e^\theta]$	$e-1-e^\theta$	

To this point we have developed these ideas under the restriction that F be analytical. However, the existence of D as a continuous function requires merely that F be differentiable once. Further, if a finite number of discontinuities in D is allowable, we may permit F to be not differentiable at a finite number of points in the θ-interval. That is, F may consist of a finite number of different analytical functions of θ, each covering a separate part of the θ-interval. The scope for using analytical methods is therefore even wider than might appear at first glance.

The range of exact solutions is so great that the $D(\theta)$ functions encountered in practical applications should be capable of accurate representation by the form (4). It would therefore seem that foundations have been provided for placing the theory and practice of much of the mathematics of concentration-dependent diffusion on an analytical basis. These matters are treated further in papers in the press in the *Australian Journal of Physics*.

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[1] Hartley, G. S., *Trans. Faraday Soc.*, **42**, B, 6 (1946).

[2] Crank, J., and Henry, M. E., *Trans. Faraday Soc.*, **45**, 636 (1949).

[3] Crank, J., and Henry, M. E., *Trans. Faraday Soc.*, **45**, 1119 (1949).

[4] Klute, A., *Soil Science*, **73**, 105 (1952).

[5] Philip, J. R., *J. Inst. Eng. Aust.*, **26**, 255 (1954).

[6] Philip, J. R., *Proc. Nat. Acad. Sci. (India)*, Allahabad, **24 A**, 93 (1955).

[7] Philip, J. R., *Soil Science*, **83**, 345, 435; **84**, 163, 257, 329 (1957); **85**, 278, 333 (1958).

[8] Philip, J. R., *Trans. Faraday Soc*, **51**, 885 (1955).

[9] Philip, J. R., *Aust. J. Physics*, **10**, 29 (1957).

[10] Crank, J., "Mathematics of Diffusion", 166 (Oxford, 1956)

[11] Fujita, H., *Text. Res. J.*, **22**, 757 (1952).

[12] Fujita, H., *Text. Res. J.*, **22**, 823 (1952).

GEOLOGY

Presence of Carbohydrates and Other Organic Compounds in Ancient Sedimentary Rocks

MOST investigators have been of the opinion that proteinaceous and carbohydrate materials that are deposited in marine and lake sediments cannot survive for long periods of time but are rapidly decomposed. However, in recent years several investigators have shown the presence of amino-acids in Tertiary shales[1] and in fossil shells and bones as old as the Devonian[2].

Preliminary studies have now demonstrated the presence of minute amounts of carbohydrate as well as proteinaceous substances in bituminous sedimentary rocks ranging in age from Ordovician to Tertiary. The reducing environment under which these sediments accumulated, coupled with the association between the organic compounds and finely divided inorganic matter, probably account for the preservation of these compounds.

Outcrop and well-core samples were thoroughly cleaned by immersion in chromic acid and subsequent washing with water. After being powdered, each sample (about 500 gm.) was treated with 0·5 *N* sulphuric acid for 8–10 hr. on a boiling water-bath. The hydrolysis mixture was neutralized with barium carbonate and freed from inorganic matter by ethanol precipitation and by ion-exchange resins. The hydrolysate from each sample was concentrated *in vacuo* and examined by paper chromatography. The following sugars were tentatively identified: glucose, xylose and arabinose, and possibly galactose and rhamnose (see Table 1). The concentration of glucose, which was the dominant component, expressed in mgm./kgm. of rock sample, varied from 0·14 for the Simpson to 1·3 for the Marcellus sample. In certain of the rock samples there was some indication of the presence of glycerol.

The glucose (180×10^6–300×10^6 years old), isolated from the Leonard and the Marcellus formations by the above treatment, was separated by paper chromatography (solvent: pyridine–ethyl acetate–water (2 : 5 : 7)) and characterized as the D-isomer by transforming it into N-*p*-nitrophenyl-β-D-glucosylamine, m.p. and mixed m.p. 184° C., $[\alpha]^{20}_{D} - 200°$ (approx.) in pyridine (*c.* 0·1)[3].

The mode of occurrence of the carbohydrate compounds in the rock formations is not yet known.

Table 1. CARBOHYDRATE COMPONENTS OF SEDIMENTARY ROCKS

Formation	Age	Component sugars				
		Glucose	Galactose	Arabinose	Xylose	Rhamnose
Stonehenge	Ordovician	+		(? +)	(? +)	
Simpson	Ordovician	+	(? +)	+	+	
Upper Chambersburg	Ordovician	+		(? +)		
Marcellus	Devonian	+	(? +)	+	+	+
Woodford	Upper-Devonian	+		+	+	
Des Moines	Pennsylvanian	+		+	+	
Missouri	Pennsylvanian	+				(? +)
Leonard	Permian	+	(? +)		+	
Schuler	Jurassic	+		+	+	
Green River	Eocene	+	(? +)		(? +)	
Elko	Miocene	+				

The rock was treated with hot 0·5 *N* sulphuric acid and the sugars identified chromatographically.

It has been shown, however, that in the case of a sample from the Marcellus formation, extraction with water yielded about 10 per cent of the total carbohydrate material as the free sugars, arabinose, glucose and xylose; the remaining undissolved carbohydrate material may be present in the polymeric form.

The amino-acids present in the acid hydrolysates of several rock samples were also investigated by paper chromatography. In comparing two core samples of shale of late Palæozoic age, the highly fossiliferous one of Pennsylvanian age appeared to contain alanine, glycine, valine, glutamic acid, aspartic acid, proline, leucine, cystine and tyrosine, whereas the non-fossiliferous shale of Permian age yielded only one amino-acid tentatively identified as aspartic acid. The amino-acids in the Pennsylvanian sample may be derived from fossil shell fragments.

Control experiments showed that neither the sugars nor the amino-acids arose from the materials used in the isolation techniques. These organic substances are believed to be original constituents of the rocks, although in the outcrop samples, despite careful collection and cleaning treatment, the possibility of surface contamination cannot be completely ruled out at present.

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[1] Erdman, J. G., Marlett, E. M., and Hanson, W. E., *Science*, 124, 1026 (1956).

[2] Abelson, P. H., "Organic Constituents of Fossils", Carnegie Inst. Washington Year Book, 53, 97 (1954).

[3] Weygand, F., Perkow, W., and Kuhner, P., *Ber.*, 84, 594 (1951).

Evidence for Tertiary Crustal Distortion in Mid-Argyll

REGIONAL deformation associated with the emplacement of Tertiary igneous rocks is well known in the British Isles. There appears to be some evidence in mid-Argyll, recorded on the one-inch maps of the Geological Survey of Scotland[1], which suggests that crustal distortion (broadly similar to that described from east Greenland by Wager and Deer[2]) may have occurred in this area during the Tertiary. This distortion is now reflected by a change in the trend of the Old Red Sandstone minor dyke swarm, the north-north-east–south-south-west tear-faults and, also, by variations in the strike of the Dalradian country-rocks; the district over which this change takes place coincides with the path of the Tertiary dyke swarm centred on Mull.

The Old Red Sandstone dyke swarm of the south-west Highlands has a north-north-east–south-south-west trend for most of its length[3]. However, just to the south of Loch Etive[1], a slight swing is seen and the dyke swarm traces out a gentle arcuate curve (Fig. 1). In the Craignish–Kilmartin area, twenty miles south-south-west of Loch Etive, the dykes have returned to their north-north-east–south-south-west trend; the greatest deflexion of the dykes from their normal orientation is about 15°. It is of importance

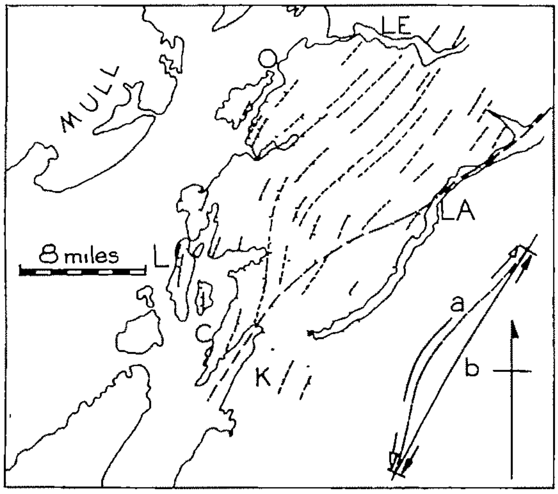

Fig. 1. Trend of Old Red Sandstone minor dykes (dashed lines) and Ericht–Craignish fault (alternately dotted and dashed line) Stippling represents the main zone of Tertiary dykes on the mid-Argyll mainland. The diagram in the lower right-hand corner illustrates the method of measurement of crustal extension (Extension = *a*–*b*/*b*). *LE*, Loch Etive; *O*, Oban, *LA*, Loch Awe; *C*, Craignish; *K*, Kilmartin; *L*, Luing

to note that the dyke swarm follows this arcuate trend, irrespective of whether the individual intrusions are emplaced into the Old Red Sandstone Lorne lavas or the Dalradian country-rocks. This same curving change in attitude is also shown by the Ericht–Laidon tear-fault which passes through Loch Awe to Loch Avich and Loch Craignish (Fig. 1); similarly, smaller tear-faults of the same set show a general arcuate swing. The strike of the Dalradian country-rocks and the axial trace of the Loch Awe syncline also appear to show the same change in trend (Fig. 2). It might be suggested that this flexure was developed in the Dalradian rocks during the Caledonian fold-movements and that the flexure then controlled the mode of emplacement of the later Old Red Sandstone dykes into both the Dalradian and the overlying Lorne lavas. This possibility appears unlikely because of the consistent north-north-east–south-south-west trend of the dyke swarm in the ground north-east of Oban[1]; in areas of Dalradian with variable strike, there is no change in trend of the dykes. Similarly, the arcuate swing in the Ericht–Craignish fault and other sub-parallel faults cannot be readily explained except by later deformation.

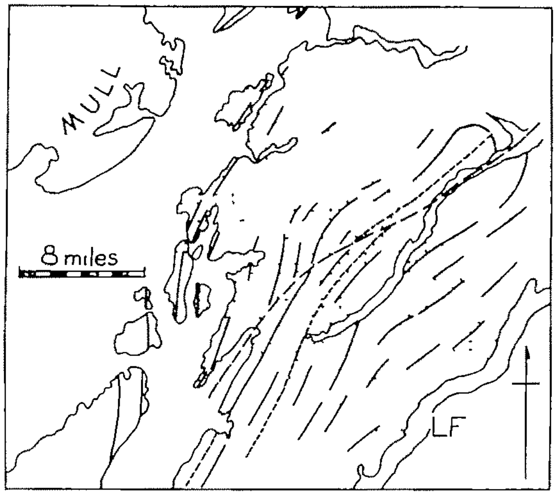

Fig. 2. Strike of the Dalradian country-rocks (solid lines) and axial trace of the Loch Awe Syncline (dashed line). Stippled area represents Mull Dyke Swarm. *LF*, Loch Fyne

It will be noted that (Figs. 1 and 2) the development of this arcuate structure is virtually restricted to the main zone occupied by the Mull Tertiary dyke swarm. The flexure appears to be dying out eastward and is scarcely recognizable near Loch Fyne (Fig. 2); there is, however, a slight curving in the Highland Boundary Fault where it is crossed by the Mull swarm, near Dunoon. Estimates of the crustal extension which might have been produced by the flexure can be made by comparing the distance along the arcuate trend of the flexure to that along the possible, undistorted trend: this is illustrated in Fig. 1. The estimate obtained in this way suggests a 3½ per cent extension between Luing and Oban and a 2½–3 per cent extension between Craignish and Loch Etive. These results can be compared to estimates, based upon actual measurements of dyke thicknesses, of a 4 per cent stretch along the south-east coast of Mull[2] and a 2½ per cent stretch along the western coast of Craignish[3].

I conclude that the coincidence in position between the arcuate flexure and the Mull dyke swarm is a strong indication that the flexure is the result of regional crustal distortion during the Tertiary.

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[1] Geol. Surv. Scotland 1-inch Sheet, Nos. 36, 37, 44, 45 and 53.

[2] Wager, L. R., and Deer, W. A., *Geol. Mag.*, 75, 39 (1938).

[3] Richey, J. R., *Trans. Edin Geol. Soc.*, 13, 393 (1939).

[4] Allison, A., *Geol. Mag.*, 73, 73 (1936).

CHEMISTRY

Chain Transfer in Mixed Solvent Systems

Chain transfer for solvents[1–4] in addition polymerization has attracted much attention, but scarcely any attempt has been made so far to determine the chain-transferring capacities of those substances which are immiscible in the monomer (say, water) or substances insoluble in the monomer, for example, such polymers as polyacrylonitrile. In the present work an attempt has been made to determine the transfer constants for such substances, keeping the whole system homogeneous in the presence of a third component.

The general equation for the reciprocal average degree of polymerization in the catalysed system can be easily derived[4] as:

$$1/\bar{P} = C_M + C_S(S)/(M) + C_I(I)/(M) + \ldots (A^2/(M)^2) \ldots R_p \quad (1)$$

where $\bar{P}$ is the number average degree of polymerization, C_M, C_S and C_I are the chain-transfer constants for monomer, solvent and initiator respectively. (S), (M) and (I) are concentrations of solvent, monomer and initiator respectively, whereas R_p is the rate of polymerization. A is a constant and is the ratio of the termination (k_t) and propagation constant (k_p); that is to say, $k^{\frac{1}{2}}_t/k_p$ for termination by coupling and $2k^{\frac{1}{2}}_t/k_p$ for termination by disproportionation. For our present purpose we have determ-

Table 1*a*

S_1, *n*-butyl alcohol ; $Cs_1 = 1\ 0 \times 10^{-5}$ (ref. 2) ; temperature, 60° C.

Solvent S_2	$M : S_1 : S_2$ (cm³)	$10^5\ R_p$ (moles/l /sec.)	$10^5\ (1/\bar{P})$	$10^5\ R_pA^2/M^2$	$10^5\ Cs_2$	
					Present work	Literature
Carbon tetrachloride	2 : 1 : 1	2 07	38·72	14·88	19 90	15·81 (ref. 2)
Cumene	2 : 1 : 1	1·22	24 67	8·76	12 60	12·48 (ref. 7)*
Ethyl benzene	2 1 : 1	1 53	25 28	10 97	10 70	9 10 (ref. 7)

Table 1*b*

S_1, Methyl ethyl ketone ; $Cs_1 = 4{\cdot}5 \times 10^{-5}$ (ref. 2)

Solvent S_2	$M : S_1 : S_2$ (cm.³)	$10^5\ R_p$ (mole/L/sec)	$10^5\ (1/\bar{P})$	$10^5\ R_pA^2/M^2$	$10^5\ Cs_2$	
					Present work	Literature
Carbon tetrachloride	1 : 1 : 1	2 46	19·95	8·08	18·30	15 81 (ref. 2)
Cumene	1 1 : 1	0 52	10 29	1 72	13·74	12·48 (ref. 7)
Ethyl benzene	1 : 1 . 1	2 02	12·72	6 66	6·72	9 10 (ref. 7)
Ethyl acetate	1 : 1 . 1	2 08	10 28	6 90	1 26	1·30 (ref. 2)
*Cyclo*hexane	1 : 1 : 1	1 18	7 10	3 90	1 00	0·85 (ref. 7)

*Ref. 7, unpublished work in this laboratory.

ined A from the slope of the plot of $1/\bar{P}$ against R_p in the polymerization of methyl methacrylate initiated by 2,2′azo*bis*-*iso*butyronitrile.

In the case where two solvents S_1 and S_2 are employed the equation may be transformed to :

$$1/P = C_M + C_{S1}(S_1)/(M) + C_{S2}(S_2)/(M) + R_pA^2/(M)^2 \quad (2)$$

If azonitriles are used as initiators the C_I of which are well known to be approximately zero, this can be further simplified to :

$$1/P = C_M + C_{S1}(S_1)/(M) + C_{S2}(S_2)/(M) + R_pA^2/(M)^2 \quad (3)$$

Now if the value of C_{S1} is known, this equation can be utilized to determine the value of C_{S2} provided the whole system is kept homogeneous. In that case :

$$C_{S2} = 1/(S_2)/(M) \ldots 1/P - C_M - C_{S1}(S_1)/(M) - R_pA^2/(M)^2 \quad (4)$$

To check the validity of equation (4) and the method proposed we have utilized a number of solvents as S_2, the chain-transfer constants of which have been determined very carefully by previous workers[3]. A number of sets of experiments were carried out with methyl methacrylate at 60° C. *n*-Butyl alcohol and methyl ethyl ketone were chosen as solvents S_1. The value of C_M has been taken from our previous work[5], and $\bar{P}$ was determined by viscometric measurements using the Ostwald type of viscometer from the relation $\bar{P} = K(\eta)^\alpha$. K and α have been taken from the work of Baxendale *et al.*[6]. Polymerization experiments were carried out by the sealing-tube method already described[2,3].

From our results represented in Table 1 it can easily be seen that the values of C_{S2} obtained by the present method compare favourably with values in the existing literature.

These results clearly show that the method suggested can be applied in evaluating chain-transfer constants for solvents. This opens up the possibilities of determining chain transfer for solvents immiscible with the monomer or in cases where the polymer separates out of the phase. Our present work relates to the chain-transferring capacity of water, the detailed results of which will be published in due course.

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[1] Mayo, F. R., *J. Amer. Chem. Soc.*, 65, 2824 (1943).
[2] Nandi, U. S., and Palit, S. R., Proc. Internat. Symp. on Macromolecules, 1954 (published in *La Reserca Scientifica*).
[3] Basu, S., Sen, J. N., and Palit, S. R., *Proc. Roy. Soc.*, A, 202, 485 (1950).
[4] Palit, S. R., Nandi, U. S., and Saha, N. G , *J Polymer Sci.*, 14, 295 (1954).
[5] Saha, N. G., Nandi, U. S., and Palit, S. R., *J. Chem. Soc.*, 7 (1958)
[6] Baxendale *et al.*, *J. Polymer Sci.*, 1, 273 (1946).

Nuclear Spin Resonance Spectrum of Isotactic Polystyrene

THE nuclear spin resonance spectrum of polystyrene has been reported by Bovey, Tiers and Filipovich[1]. Resonance peaks for *ortho* and for *meta* and *para* aromatic protons were observed. Absorptions by methylene and carbinyl (chain) protons appeared as a broader single peak in the case of high molecular weight species. For polystyrene of degree of polymerization 10 or less the carbinyl proton was resolved from that of the methylene protons.

We have now observed that carbinyl and methylene proton peaks are partially resolved also for high

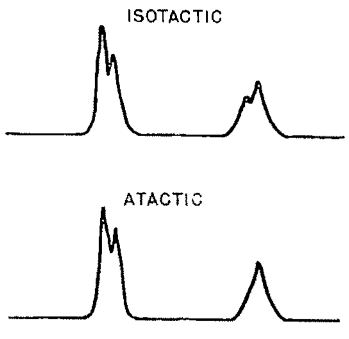

Fig. 1

molecular weight (10^5–10^6) isotactic polystyrene. The spectrum shown was obtained from isotactic polystyrene in deuterated chloroform using a 'Varian *V*–4300' 40·00-Mc./s. spectrometer. A similar spectrum was obtained in thionyl chloride. Like Bovey *et al.*, we observed no resolution of chain protons for commercial atactic polystyrene. Neither was resolution observed for high molecular weight (10^5) polystyrene prepared using *n*-butyl-lithium initiator nor for benzene-insoluble polystyrene obtained via Alfin sodium alkyl catalysis[2,3].

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[1] Bovey, F. A., Tiers, G. V., and Filipovich, G., *J. Polymer Sci.*, **38**, 73 (1959).

[2] Morton, A. A., and Grovenstein, E., *J. Amer. Chem. Soc.*, **74**, 5453 (1952).

[3] Williams, J. R., Vandenberge, J., Dunham, K. R., and Dulmage, W. J., *J. Amer. Chem. Soc.*, **79**, 1716 (1957).

Dissolution of Chrysotile Asbestos in Water, Acid and Alkali

The dissolution of the asbestos mineral, chrysotile, $(OH)_6Mg_6Si_4O_{11}$, has been studied as a part of investigations into the nature of the industrial lung disease, asbestosis[1]. Because the results may be of value to industrial chemists and to geologists, and they explain apparently anomalous results which have been published[2,3], they are outlined here although the details and a discussion on their relevance to the problem of pneumoconiosis will appear elsewhere.

At 85–90° C. chrysotile gives an extract containing appreciable amounts of monosilicic acid and magnesium, for example, 0·5 gm. chrysotile in 100 ml. water gives 0·4 mgm. magnesium and 0·2 mgm. *ortho*silicic acid per 100 ml. solution after 15 min. When extracted with water at 25° C. for some days, chrysotile gives a very low concentration of silicic acid in solution but more than ten times that concentration of magnesium. If the asbestos is removed and the extract is heated to 85–90° C., the concentration of the dissolved silicic acid increases more than ten-fold although the magnesium concentration is unaffected. As an example, after 5 days, 1 gm. chrysotile in 200 ml. water at 35° gave an extract containing 1·1 mgm. magnesium and 0·1 mgm. *ortho*silicic acid per 100 ml. After heating for 3 hr. at 90° C. the extract contained 1·2 mgm. magnesium and 1·4 mgm. *ortho*silicic acid per 100 ml. It is thought that magnesium ions are leached from the chrysotile giving fragments of the two-dimensional sheets of the silicon–oxygen lattice which are resistant to water except at elevated temperatures, when they break down to give *ortho*silicic acid.

Hydrochloric acid (0·1 *N*) attacks chrysotile giving a solution containing *ortho*silicic acid and magnesium ions. Sodium hydroxide (0·1 *N*) scarcely attacks chrysotile, presumably because of a protective coating of hydrated magnesium ions on the surface of the fibres. If acid-washed chrysotile is extracted with sodium hydroxide, silica passes rapidly into solution but very little magnesium; the acid removes the protective layer, allowing the disintegration of the next layer of the silicon–oxygen lattice by the alkali. Evidently the behaviour of chrysotile depends on its history, and it is particularly vulnerable to alternating acid and alkaline conditions.

This communication describes a part of a programme of research which is proceeding under the auspices of the Asbestosis Research Council.

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[1] Holt, P. F., "Pneumoconiosis",142 (Arnold, London, 1957).

[2] Briscoe, H. V. A., Matthews, J. W., Holt, P. F., and Sanderson, P. M., *Trans. Inst. Min. and Metall.*, **44**, 291 (1937).

[3] King, E. J., and McGeorge, M., *Biochem. J.*, **32**, 417 (1938).

Clathrate Compounds in Chromatography

Partition and adsorption chromatography can be successfully applied for separation of mixtures of substances which differ in type and number of functional groups, or for separation of members of a homologous series. Less success was attained in separation of isomers. We therefore sought for a process which would be selective as regards the shape and size rather than the chemical properties of molecules.

The discovery of clathrate compounds suggested that inside their structure molecules of definite shape and size might be incorporated[1,2]. The first inorganic compound of that group was monoamminonickel cyanide[3,4], but many other complexes[5] of nickel, cobalt, manganese and iron with four basic nitrogen molecules and some anions around the metal ion followed. These compounds are said to be highly selective towards clathrate formation with organic molecules; and, more important, their selectivity depends rather on the shape of incorporated molecules than on their volume.

The selectivity, especially towards aromatic molecules, the small solubility of some complexes of that type, and other properties suggested the use of the formation of clathrate compounds for the separation of substances on the chromatographic column.

In preliminary experiments we filled the column with water-insoluble tetra-(4-methylpyridino)-nickel dithiocyanate prepared by the method of Schaeffer *et al.*[5]. Later, we found that another complex, prepared by using for precipitation a mixture of β- and γ-picoline and 2,6-lutidine instead of pure γ-picoline (so-called 3rd coal tar base fraction), has better properties.

One of the most important problems was to find an appropriate solvent composition for use as a mobile phase. We have used the mixture of γ-picoline or 3rd fraction with water containing some potassium thiocyanate.

To obtain high precision of results, we have used the chromato-polarographic method[6]. The compounds separated and incorporated were aromatic and aliphatic nitro-compounds.

In these conditions we have investigated the separation of isomeric nitrophenols, nitroanilines, chloronitrobenzenes and nitrotoluenes. In all instances quantitative separation of *ortho* from other isomers was obtained. The *ortho* isomers are found in the first fractions of eluate and can be very accurately determined polarographically. Using more concentrated solutions of potassium thiocyanate and 3rd fraction, *meta*- and *para*-isomers can be eluted. In some instances the mobile phase composition can be so chosen that it is possible to separate completely all three isomers in one process. We have used 2·5 *M* potassium thiocyanate in 10 per cent 3rd fraction

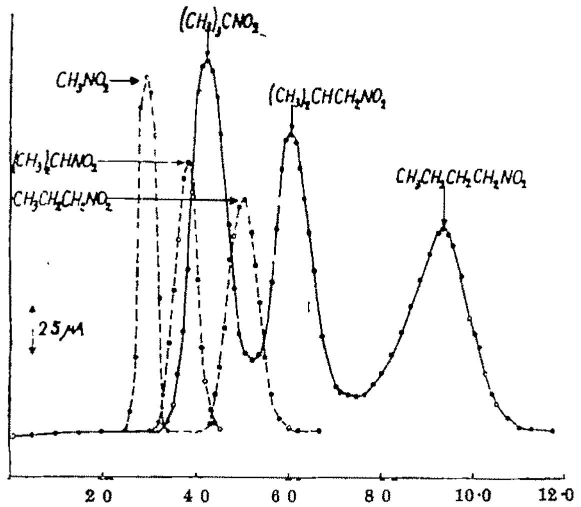

Fig. 1

base mixture for the separation of isomeric nitrophenols and nitroanilines (Kemula, W., Sybilska, D., and Geisler, J., unpublished work).

From nitroaliphatic compounds we have studied mixtures of 1-nitropropane and 2-nitropropane, 1-nitrobutane, 2-methyl-1-nitropropane and 2-methyl-2-nitropropane, but their separation is more difficult. No separation occurs when columns were filled with tetra-(4-methylpyridino)-nickel dithiocyanate or tetra-(3-methylpyridino)-nickel dithiocyanate. No insoluble complex could be prepared from pure 2,6-lutidine, but using the 3rd fraction base a very satisfactory column filling was obtained.

In Fig. 1 the continuous line indicates the separation of nitrobutanes, the dashed line indicates the elution of nitromethane and nitropropanes. The column height was 17 cm., its inner diameter 5·8 mm.; as an eluant we used 0·25 *M* potassium thiocyanate in 2 per cent 3rd fraction solution in water.

In Fig. 2 is shown for comparison the separation of the isomers using the reverse phase partition chromatography on powdered rubber[7]. As a stationary phase we used *n*-heptane, as a mobile phase 0·25 *M* potassium thiocyanate in 2 per cent 3rd fraction base mixture in water. The column height was 21·5 cm., the diameter 5·8 mm.

Our results show that the application of clathrate compounds in chromatography affords an excellent method for the separation of isomers. The mode of action of the complex prepared from 3rd fraction base mixture and the applicability of other complexes are being investigated. Such chromatographic studies may also be helpful in elucidating the properties of clathrate compounds and their structure.

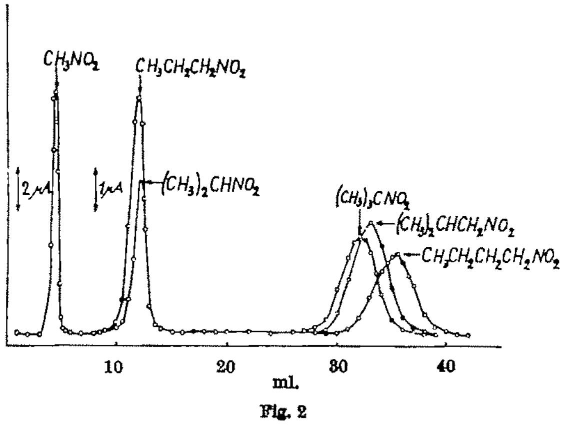

Fig. 2

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[1] Powell, H. M., *J. Chem. Soc.*, 61 (1948).
[2] Kramer, F. D., *Rev. Pure App. Chem.*, 5, 143 (1955).
[3] Hofman, K. A., *Z. anorg. Chem.*, 15, 204 (1897).
[4] Powell, H. M., and Rayner, J. H., *Nature*, 163, 566 (1949).
[5] Schaeffer, W. D., Dorsey, W. S., Skinner, D. A., and Christian, C. G., *J. Amer. Chem. Soc.*, 71, 5870 (1957).
[6] Kemula, W., *Roczniki Chem.*, 26, 281 (1952); *Przemysl Chem.*, 33, 453 (1954).
[7] Kemula, W., and Sybilska, D., *Chemia Anal.* (*Warsaw*), 4, 123 (1959).

BIOCHEMISTRY

Casein labelled with Iodine-131 as a Substrate for the Measurement of Trypsin and Chymotrypsin

THE spectrophotometric method of Kunitz[1] is a marked improvement over previous methods for the measurement of trypsin and chymotrypsin activity. This procedure involves a measurement of the optical density at 280 mμ of the trichloracetic acid-soluble products formed during the enzyme hydrolysis of casein; it becomes impractical, however, where large concentrations of 280 mμ absorbing materials are present, such as in sera. In this communication, a modification of this procedure will be described which utilizes casein labelled with iodine-131 as the substrate and the measurement of radioactivity as an estimate of enzyme activity.

Labelled casein is prepared in the following manner. Into 12 ml. of 0·1 *M* disodium hydrogen phosphate–potassium dihydrogen phosphate buffer (*p*H 7·60) is placed 1 gm. of Hammarsten casein (Hammersten casein from Nutritional Biochemical Corporation); trypsin (50 per cent magnesium sulphate, 2 × crystallized) and chymotrypsin (salt-free, crystallized) from Worthington Biochemical Corporation; radioactive iodine from Abbott Laboratories, specify chemically pure product). The protein is stirred into solution while heating in a warm water-bath and then heated for 15 min. in a boiling water-bath. After cooling the solution to room temperature, the flask is placed on a magnetic stirrer and the casein iodinated with the following solution. Into 1 ml. of a sodium iodide solution containing 1 mc. of iodine-131 is pipetted the following reagents: 0·3 ml. 0·01 *M* potassium iodate, 0·3 ml. 0·01 *M* potassium iodate, drop by drop 0·05 *M* hydrochloric acid until the sodium iodide–iodine-131 mixture turns faint yellow, followed by 0·5 ml. of 0·05 *M* hydrochloric acid. The orange-coloured radioactive solution is mixed thoroughly and then added dropwise to the stirring casein solution over a period of 10–15 min. Stirring is maintained at room temperature for an additional 45 min. The iodinated casein is then transferred to a dialysis bag and dialysed against four 700-ml. portions of 0·1 *M* phosphate buffer (*p*H 7·35) in the refrigerator. The resulting labelled casein has a specific activity of 15–20 μc./ml. for the final casein solution and constitutes a 20–25 per cent radiometric yield.

The specific activity of free iodine in the fourth dialysate, which is also representative of the free iodine-131 contaminate of the labelled casein, is only on the average of 1·0 mμc./ml. This constitutes approximately 0·005 per cent of the total iodine-131 in the final casein solution.

Stock casein, unlabelled and un-iodinated, is used for making the appropriate dilution of the labelled substrate and is prepared as above. For enzyme measurement, a 2 per cent casein solution containing 1 μc. per ml. is prepared in 0·1 *M* phosphate buffer (*p*H 7·35).

Stock solutions of trypsin and chymotrypsin are prepared as 250 μgm./ml. in 0·0025 *M* hydrochloric acid. Diluted solutions of 25 μgm./ml. in 0·0025 *M* hydrochloric acid are further prepared for actual use.

Enzyme measurement is carried out as follows: Into 12-ml. centrifuge tubes are pipetted varying amounts of trypsin or chymotrypsin contained in less than 1 ml. volume. The volumes are made up to 1·0 ml. with 0·0025 *M* hydrochloric acid, mixed, and pre-incubated in a 37·5° C. constant-temperature bath for 5 min. The 2 per cent casein solution labelled with iodine-131 is pre-incubated at the same time. 1-ml. volumes of the casein solution are added with stirring to the assay tubes at 1-min. intervals. A blank tube containing 1 ml. of 0·0025 *M* hydrochloric acid is treated similarly. After a 20-min. incubation period, 3 ml. of 5 per cent trichloracetic acid are added with stirring to stop the enzyme action. The stirring rods are carefully removed and the solutions left at room temperature for 1 hr. Following a 20-min. centrifugation at 1,700*g* and 25° C., the supernatants are poured from the tubes. 1-ml. volumes of the supernatants are pipetted into 1-dram, screw cap vials and the radioactivity therein measured in a well scintillation detector.

In Fig. 1 is shown a typical plot of corrected counts (corrected for coincidence loss and for counts obtained in the blank tube) versus μgm. trypsin per ml. of the 1 per cent casein digestion mixture. Measurements of the radioactivity in the supernatants have been found to correlate well with optical density measurements made at 280 mμ, as indicated in Fig. 1. Fig. 2 shows a similar curve obtained for the measurement of chymotrypsin activity by this method.

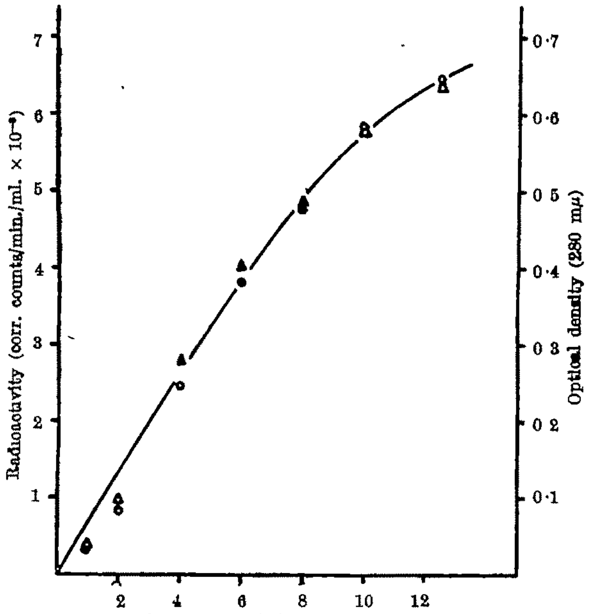

Fig. 1. Standard curve for digestion of casein labelled with iodine-131 by trypsin. Counts/min./ml (circles) in trichloracetic acid supernatant and/or optical density (triangles) plotted versus micrograms of trypsin protein

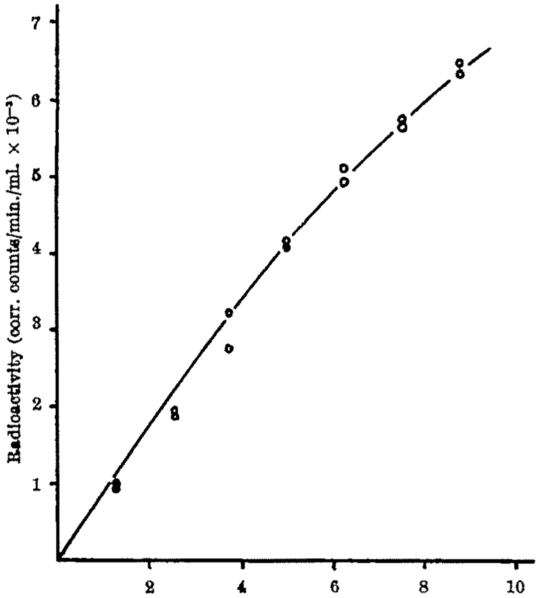

Fig. 2. Standard curve for digestion of casein labelled with iodine-131 by chymotrypsin. Counts/min./ml. in trichloracetic acid supernatant plotted versus micrograms of chymotrypsin protein

The use of a 2 per cent casein solution provides a linear relationship between enzyme activity and enzyme concentration over a greater range of enzyme concentration than is possible with the 1 per cent casein used by Kunitz[1]. Both 3 per cent and 4 per cent solutions serve to extend this linear relationship over a still further range of enzyme concentration. However, when using these more concentrated casein solutions, a relatively large amount of precipitate is obtained upon treatment with trichloracetic acid. Therefore, a correction should be made for the precipitates associated with each tube to obtain the correct curve representing enzyme activity.

This procedure utilizing labelled casein provides a sensitive means of measuring trypsin or chymotrypsin activity. Only those casein hydrolysis products which result directly from enzyme activity are measured. The spectrophotometric method could conceivably measure acid-soluble contaminants which absorb highly in the region of 280 mμ in addition to the casein hydrolysis products.

This method has been applied to the evaluation of the trypsin inhibitory capacity of sera of normal individuals and of patients in varying diseased states which are in part associated with altered trypsin metabolism.

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[1] Kunitz, M., *J. Gen. Physiol.*, 30, 291 (1947).

Role of the Inducer in the Induced Synthesis of β-Galactosidase in *Escherichia coli*

INDUCED (adaptive) enzyme formation in micro-organisms has been the subject of numerous studies[1]. Although a great deal is known about the kinetics of the biochemical processes involved, the precise role of the inducer remains enigmatic.

Any hypothesis describing the mode of action of the inducer must account for the structural complementariness between the inducer and the enzyme elicited by the inducer. Yudkin's 'mass action hypothesis'[2], which postulates inducer-enzyme combination as the prime mover of induction, represents such an attempt. The mass action hypothesis implies that substances capable of inducing an enzyme have affinity for the enzyme. Furthermore, the hypothesis predicts a positive correlation between the relative affinity of an inducer for the induced enzyme and the efficacy of the inducer as inducer of the enzyme. These predictions have been tested employing several inducible enzymes and a number of inducers in bacteria and yeasts[3]. It was found that certain highly effective inducers had no, or low, affinity for the induced enzyme. For this reason and also on the basis of certain other findings apparently incompatible with a mass action mechanism of enzyme induction, it was suggested that the inducer exerts its effect not by combining with the enzyme directly but rather by combination with some other cellular constituent. This hypothetical inducer-cell complex, the 'organizer'[4,5], would then catalyse formation of the enzyme.

One of the most thoroughly analysed inducible enzyme systems is that of the β-galactosidase of *Escherichia coli*. Monod and his collaborators[6,7] in a study of the relationship between the affinity of non-metabolizable galactosides for the enzyme β-galactosidase and the effectiveness of the same galactosides as inducers of the enzyme found that thiomethyl-β-D-galactoside and the α-galactoside melibiose were both highly effective inducers of β-galactosidase but had very low affinity for the enzyme. Affinity for β-galactosidase was determined by measuring the inhibition of the hydrolysis of the test substrate *o*-nitrophenyl-β-D-galactoside by the other two galactosides. A cell-free enzyme preparation was employed. It has been shown, however, that cell-free β-galactosidase (or the β-galactosidase of bacteria treated with toluene) differs from the intact-cell β-galactosidase with respect to its kinetic behaviour[8]. It has also been found more recently[9] that concomitant with β-galactosidase synthesis, the induced bacteria develop the ability to concentrate galactosides intracellularly. In view of these findings it was decided to re-examine the relationship between the affinity of galactosides for β-galactosidase and their inducing efficacy by (*a*) measuring the ability of galactosides to inhibit the β-galactosidase activity of intact cells and (*b*) determining the inducing efficacy of the same galactosides in *E. coli* strain *ML* 3, a strain that is unable to accumulate galactosides[9], and in which galactosides enter the cell apparently by diffusion only[10].

Fig. 1 shows that thiomethyl-β-D-galactoside and melibiose inhibit intact-cell β-galactosidase activity (measured as *o*-nitrophenyl-β-D-galactoside hydrolysis) competitively. Intact-cell β-galactosidase activity was measured in *E. coli* strain *ML* 308, which forms β-galactosidase constitutively and also has the ability to concentrate galactosides intracellularly[9]. The reaction was carried out as described in an earlier publication[11]. Michaelis constants of K_s $1 \cdot 3 \times 10^{-3}$ M *o*-nitrophenyl-β-D-galactoside and K_i $3 \cdot 3 \times 10^{-3}$ M thiomethyl-β-D-galactoside and $2 \cdot 5 \times 10^{-4}$ M melibiose were obtained. Thiomethyl-β-D-galactoside and melibiose do then show considerable affinity for the β-galactosidase of intact cells.

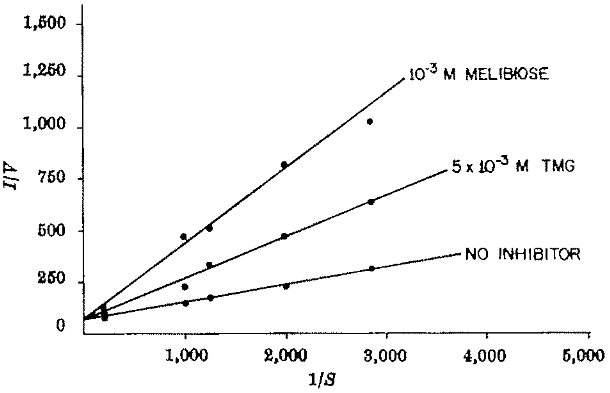

Fig. 1. Competitive inhibition of intact-cell β-galactosidase by thiomethyl-β-D-galactoside (TMG) and melibiose. Abscissa, reciprocal of *o*-nitrophenyl-β-D-galactoside concentration; ordinate, reciprocal of intact-cell β-galactosidase activity

The affinity of an inducer for the hypothetical enzyme-forming system may also be expressed as a constant $K_{s\,\mathrm{ind.}}$, where the latter term denotes that concentration of inducer which will induce a cellular concentration of enzyme that corresponds to half the maximal value obtainable, employing a saturating concentration of the same inducer and identical growth conditions. The $K_{s\,\mathrm{ind.}}$ for thiomethyl-β-D-galactoside and melibiose were determined in *E. coli* strain *ML* 3 by growing the bacteria from an inoculum of approximately 10^6 bacteria per ml. to a final population of approximately 10^9 bacteria per ml. in the presence of a series of inducer concentrations and determining the specific β-galactosidase activities of the cultures. Fig. 2 shows the $K_{s\,\mathrm{ind.}}$ for thiomethyl-β-D-galactoside to be $3 \cdot 5 \times 10^{-3}$ M and that for melibiose to be 2×10^{-4} M.

The finding that the affinities of thiomethyl-β-D-galactoside and melibiose for the enzyme β-galactosidase, when measured in the intact cell, are the same

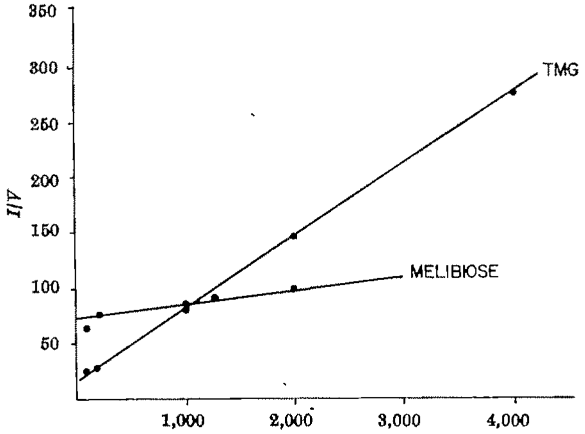

Fig. 2. Relation between the concentration of inducer present during growth and the specific β-galactosidase activity of the culture. Abscissa, reciprocal of inducer concentration; ordinate, reciprocal of specific β-galactosidase activity

as the affinities of thiomethyl-β-D-galactoside and melibiose for the enzyme-forming system is compatible with the hypothesis that enzyme induction involves combination between the inducer and the enzyme itself. The simplest scheme that would explain induced enzyme formation on this basis might be formulated thus:

$$X \text{ — enzyme} + \text{inducer} \rightarrow X + \text{enzyme — inducer}$$
$$\text{enzyme — inducer} \rightleftharpoons \text{enzyme} + \text{inducer}$$

where X denotes the enzyme-forming system. Combination of the enzyme with the enzyme-forming system X in the absence of inducer might then constitute 'repression' of enzyme formation[12].

An amplification of the proposed hypothesis of induced enzyme formation, an attempt to relate the phenomenon of inducible galactoside accumulation to the induction of β-galactosidase, as well as further experimental data bearing on the hypothesis will be published elsewhere.

I am indebted to the National Science Foundation for a research grant and to Mrs. Helen M. Talbott for expert technical assistance.

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[1] Pollock, M. R., "Induced Formation of Enzymes", in "The Enzymes", ed. by Boyer, B. P., Lardy, H., and Myrbäck, K., (Academic Press, Inc., New York, 1959).

[2] Yudkin, J., *Biol. Rev.*, **13**, 93 (1938).

[3] Mandelstam, J., *Int. Rev. Cytol.*, **5**, 51 (1956).

[4] Pollock, M. R., *Brit. J. Exp. Path.*, **31**, 739 (1950).

[5] Cohn, M., and Monod, J., in "Adaptation in Microorganisms", 132 (Cambridge Univ. Press, London, 1953).

[6] Monod, J., in Jessup Lectures delivered at Columbia University (1954).

[7] Monod, J., Cohen-Bazire, G., and Cohn, M., *Biochim. Biophys. Acta*, **7**, 585 (1951).

[8] Lederberg, J., *J. Bact.*, **60**, 381 (1950).

[9] Rickenberg, H. V., Cohen, G. N., Buttin, G., and Monod, J., *Ann. Inst. Past.*, 91, 829 (1956).

[10] Herzenberg, L. A., *Biochim. Biophys. Acta*, **31**, 525 (1959).

[11] Rickenberg, H. V., Yanofsky, C., and Bonner, D. M., *J. Bact.*, **66**, 683 (1953).

[12] Vogel, H. J., *Proc. U.S Nat Acad. Sci.*, **43**, 491 (1957).

Ester-like Groups in Different Collagens and Gelatins

Gallop *et al.*[1] have recently reported the presence of ester-like links in several soluble collagens. We had also found such links in commercial acid-precursor pigskin gelatin and bovine hide corium collagen.

Gelatin was treated with 1·5 M hydroxylamine at pH 8·6 in water, 5 M or 8 M urea or 5 M lithium bromide at 4° C. for 1 or 5 days or at room temperature for 4 hr. Bovine hide corium collagen (prepared by washing with water and 8 per cent sodium chloride and acetone) was first soaked in 4 M sodium perchlorate for 24 hr., the supernatant filtered off and the residue soaked in 3 M hydroxylamine (25 ml./gm. of collagen) at pH 8·65 for 16 hr. at room temperature. Almost all the collagen dissolved except for a few small shreds. The solution was dialysed at 4° C. against water with mixed ion-exchange resins in the external compartment. The protein solution gelled. 80 per cent of the original collagen was obtained as gelatin.

Analyses for hydroxamate were carried out on the pigskin product by the ferric hydroxamate method of Lipmann and Tuttle[2] (acethydroxamic acid as standard) and by the method of Bergmann and Segal[3]. The bovine collagen was analysed by the latter method.

The hydroxamate content of the acid pigskin gelatin varied from 0·5 to 1·5 moles/10^5 gm. The results were the same for all the methods of treatment mentioned above. The hide collagen had a hydroxamate content of 2 moles/10^5 gm.

The starting pigskin gelatin has a number average molecular weight of 100,000 [4]. If one ester-like group were broken to yield two fragments of random size the average molecular weight would be 50,000. From measurements of intrinsic viscosity molecular weights of 25,000–35,000 were found, based on a viscosity–molecular weight relation established for this gelatin[4]. From melting points of 5 per cent gels, molecular weights of 30,000–70,000 were obtained, based on a melting point–molecular weight relation[4]. The discrepancy between the molecular weights by viscosity and melting point methods suggests that the reaction with hydroxylamine alters the shape of the gelatin molecules or changes the molecular weight distribution.

Hydroxamate values of 1–2 moles/10^5 gm. could be accounted for by carbohydrate; as little as 0·1 per cent would suffice. The differences between these values and the value of 10 moles/10^5 gm. found by Gallop *et al.* for soluble collagens suggest there may be differences related to whether the soluble or 'insoluble' collagen is used. There may also be differences resulting from species, tissue of origin and age of the collagen or the organism. Ageing organisms may contain increasing numbers of ester-like cross-links.

If water or sodium sulphate is used in place of sodium perchlorate, the collagen does not dissolve in hydroxylamine. Strong solutions of urea or sodium thiocyanate can be used in place of sodium perchlorate.

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[1] Gallop, P. M., Seifter, S., and Meilman, E., *Nature*, **183**, 1659 (1959).

[2] Lipmann, F., and Tuttle, C., *J. Biol. Chem.*, **159**, 21 (1945).

[3] Bergmann, F., and Segal, R., *Biochem. J.*, **62**, 542 (1956).

[4] Bello, J., Bello, H. R., and Vinograd, J. R. (to be published).

Biosynthesis of Ergot Alkaloids

Five schemes for biosynthesis of the lysergic acid moiety of the ergot alkaloids have been proposed recently. All of them involve tryptophan or 5-hydroxytryptophan as precursors[1,2].

This communication deals with the effect of tryptophan, 5-hydroxytryptophan and its related compounds on the yield of total ergot alkaloids utilizing an *in vitro* culture of *Claviceps purpurea* to determine whether experimental evidence could be obtained to support one of the above schemes. A strain of *Claviceps purpurea*, *P.R.L.* 1578, was employed using Abe's medium[3].

Inoculation of the culture and the estimation of total alkaloids were carried out by a procedure slightly modified from that of Taber and Vining[4], and the composition of the total alkaloids was established by paper chromatography (ref. 5 and Weir, J. R., unpublished work). The R_F values of the spots found

Table 1. THE EFFECT OF TRYPTOPHAN AND 5-HYDROXYTRYPTOPHAN ON THE YIELD OF ALKALOID PRODUCTION

Age of cultures (day) Feeding	Age of cultures (day) Harvesting	Additives	Mgm./l.	Mgm. alkaloid/l. Supplemented	Mgm. alkaloid/l. Control	Deviation (per cent)
11	23	DL-tryptophan	48·4	0·21	0 25	−16
11	36			2·77	1·99	+39
11	23	DL-5-hydroxy-tryptophan	32·8	0·21	0·25	−16
11	36			0·99	1·99	−50
22	29	DL-tryptophan	48·4	0·65	0·97	−38
22	43			2·17	3·27	−33
22	29	DL-5-hydroxy-tryptophan	32·8	0·61	0·97	−38
22	43			2·42	3·27	−26
30	51	DL-tryptophan	96·8	6·92	4 29	+61
30	51		242·0	6·60	4·29	+54
30	51	DL-5-hydroxy-tryptophan	90·4	5·75	4·29	+34
30	51		226·0	5 07	4·29	+18

were identical with the R_F values of pure ergosine, ergosinine or ergotaminine, ergokryptinine, elymoclavine.

Tryptophan, 5-hydroxytryptophan, indole, 5-hydroxyindole, serotonin were added to the cultures of different ages (11, 22, 30 days) and sampled after certain intervals (23, 29, 36, 43, 51 days).

Table 1 shows that addition of tryptophan or 5-hydroxytryptophan did not change significantly the yield of total alkaloids even if the amount of added DL-tryptophan or DL-5-hydroxytryptophan was gradually increased. An equivalent amount of indole, 5-hydroxyindole and serotonin also did not cause marked difference in the total alkaloid production.

In another experiment a replacement culture technique was used. On the thirty-ninth day, when alkaloid formation was 1·2 mgm./l. in the control culture, the growth medium was decanted off, the remaining mycelium was washed thoroughly with sterile water (3 × 20 ml.) and the following new media were added under sterile conditions: (*a*) 1 gm. mannitol in 20 ml. water; (*b*) 1 gm. mannitol, 0·160 gm. succinic acid in 20 ml. water; (*c*) 1 gm. mannitol, 0·160 gm. succinic acid, 0·0048 gm. DL-tryptophan in 20 ml. water; (*d*) 1 gm. mannitol, 0·160 gm. succinic acid, 0·0045 gm. DL-5-hydroxytryptophan in 20 ml. water; (*e*) 1 gm. mannitol, 0·160 gm. succinic acid, 0·0048 gm. DL-tryptophan, 0·0045 gm. L-proline, 0·0045 gm. L-valine, 0·0045 gm. L-leucine in 20 ml. water; (*f*) 1 gm. mannitol, 0·160 gm. succinic acid, 0·0045 gm. DL-5-hydroxytryptophan, 0·0045 gm. L-proline, 0·0045 gm. L-valine, 0·0045 gm. L-leucine in 20 ml. water.

After an incubation period of 19 days the total alkaloids were estimated. The results given in Table 2 indicate that there is no marked difference in the amount of total alkaloids in the replacement cultures and the cultures grown in the Abe's media harvested on the same day (control culture).

These experiments emphasize the significant role of the age of mycelium on the amount of alkaloid production of *Claviceps purpurea*. This observation seems to be supported by another experiment, when the medium of the 21-day-old culture (at this time no measurable amount of alkaloid was yet formed in the control culture) was inoculated with fresh mycelium. After harvesting the culture on the fifty-first day, the amount of total alkaloid was negligible although the weight of the mycelium was as usual.

Table 2. THE EFFECT OF TRYPTOPHAN, 5-HYDROXYTRYPTOPHAN ON THE YIELD OF TOTAL ALKALOID PRODUCTION IN REPLACEMENT CULTURES

Medium	Mgm. alkaloid/l. Replacement culture	Mgm. alkaloid/l. Control culture
a	4·61	5·07
b	4·28	5·07
c	4·47	5·07
d	4·54	5·07
e	4·86	5·07
f	5·11	5·07

Although all our experiments indicate that tryptophan and 5-hydroxytryptophan are not directly concerned in the biosynthesis of lysergic acid alkaloids, it cannot be concluded that these amino-acids are not the direct or indirect precursors of lysergic acid alkaloids. It is possible that the metabolism of these amino-acids is much faster in other directions than in the formation of alkaloids; therefore, they have no significant influence on the yield of alkaloid.

While this work was in progress, Suhadolnik *et al.*[6] reported that labelled DL-tryptophan was not incorporated into ergonovine produced by *Claviceps purpurea* cultured on rye. More recently, Taber and Vining[7] reported that large amounts of L-tryptophan (500 mgm./l. culture medium) doubled the amount of total ergot alkaloids formed by *Claviceps purpurea in vitro*. These authors also observed that the addition of L-tryptophan resulted in the production of new types of alkaloids and tryptamine. A direct comparison of the results obtained by Suhadolnik *et al.* and Taber and Vining with ours is difficult due to the different experimental conditions. (During the preparation of this communication an abstract of a paper appearing in the Bacteriological Proceedings, Fifty-ninth General Meeting, St. Louis, Missouri, was kindly submitted by W. A. Taber and L. C. Vining. Although no detailed results were given in this abstract it was stated that a culture of *C. purpurea* when supplied with tagged DL-tryptophan produced labelled lysergic acid derivatives.)

Use of the sensitive radioactive tracer technique is definitely indicated to obtain more specific information on the biogenesis of these alkaloids. This work is in progress and will be published in the near future.

This work was supported by a grant, *A*-686, from the National Research Council of Canada. We wish to express our appreciation to Dr. A. Hofmann, Sandoz, Ltd., Basle, for samples of the pure alkaloids, and to Dr. W. A. Taber and Dr. L. C. Vining, Prairie Regional Laboratory, Saskatoon, Saskatchewan, for the culture *P.R.L.* 1578.

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[1] Saxton, J. E., *Quart. Rev.*, 10, 108 (1956).
[2] Feldstein, A., *Experientia*, 12, 475 (1956).
[3] Abe, M., *J. Agric. Chem. Soc.*, Japan, 22, 2 (1948).
[4] Taber, W. A., and Vining, L. C., *Can. J. Microbiol.*, 3, 1 (1957).
[5] Taber, W. A., and Vining, L. C., *Can. J. Microbiol.*, 3, 55 (1957).
[6] Suhadolnik, R. J., Henderson, L. M., Hanson, J. B., and Loo, Y. H., *J. Amer. Chem. Soc.*, 80, 3153 (1958).
[7] Taber, W. A., and Vining, L. C., *Canad. J. Microbiol.*, 4, 612 (1958).

Glacial Acetic and Picric Acids as a Fixation for Gel Precipitation

DIFFICULTIES are often encountered in reading off immunological reactions on gel plates (cf. immunoelectrophoresis). The gel method, however, has many indisputable advantages over tube precipitation even if the latter is considered more sensitive quantitatively. The former has often been utilized for qualitative studies of the various components involved in immunological reactions. In this connexion it is often important to know whether or not the plate contains a precipitate which is not visible either macroscopically or microscopically (cf. interference microscopy). Various fixatives have been employed in order to detect or accentuate an immunological reaction before proceeding to qualitative studies. Acetic acid and alcohol in various concentrations have been most commonly used for this purpose.

During work on an immunological system in which one could *a priori* expect a very weak precipitation reaction several fixatives were tested. These were chosen with consideration of their physical and chemical effects upon the various components in the system. It appeared that a combination of glacial acetic acid and saturated picric acid solution was preferable in those instances in which a quantitative reading of a precipitation reaction involving mainly proteins is desired.

The most satisfactory combination was found to be 6 per cent glacial acetic acid and 94 per cent saturated picric acid solution ; $p\text{H} = 2 \cdot 1$. In these proportions the penetrating effect of acetic acid and the protein-precipitating effect of picric acid appear to advantage. These acids seem to complement each other in other respects as well. (At this *p*H the protein-precipitating effect of picric acid is good and *p*H is not appreciably altered by the precipitating reaction.)

If this method is to be used when serum forms one of the immunological components, the serum must be highly diluted. If this is not done, the gel plate must be washed before being fixed (cf. agar gel with serum added) otherwise interpretation is made impossible by the large amount of precipitate originating from the other serum proteins.

In addition to its sensitivity, the method described here offers other advantages such as a short fixation time and a result which can easily be interpreted and recorded.

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Scanning of Paper Electrophoretograms after Protein Dyeing with Bromocresol Green

THE blue colour of bromocresol green-dyed protein developed by exposure to ammonia vapour is not stable, and elution of the dyed protein has been used to obtain an electrophoretic curve from paper electrophoretograms of serum dyed with bromocresol green. One disadvantage of this is that the paper strip is destroyed when it would have been of value as a record for reference.

The use of an agent other than ammonia vapour was investigated with the view of stabilizing the blue colour of such dyed protein so that scanning could be carried out.

An organic base, with which the strip could be easily impregnated, strong enough to develop the blue colour of bromocresol green and which was miscible with the scanning oil, was looked for. Of several which were tried diethylamine was found to be suitable and the following method of use was found satisfactory.

The electrophoretograms were dried and then dyed according to the method described by Franglen and Martin[1]. The dry strips were then dipped in a solution of 10 per cent v/v solution of diethylamine in diethyl ether. The ether was allowed to evaporate and the blue colour of the bromocresol green-dyed protein developed. A series of dyed strips can be brought to this stage ready for scanning. The strips were then oiled for scanning using the mixture described by King and Wootton[2].

Scanning was carried out using an *E.E.L.* scanner with an Ilford 205 filter. After scanning, the oil and diethylamine were removed from the strips with either a 1 : 1 mixture of acetone and diethyl ether or benzene. The acetone used must be free from water otherwise diffusion of the dyed protein bands will occur. The strips were then air-dried ready for storage.

It was found that the blue colour remained stable for several weeks. This was probably due to the oil and diethylamine being removed by a non-ionic solvent leaving the bromocresol green in its blue form.

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[1] Franglen, G. T., and Martin, N. H., *Biochem. J.*, 57, 626 (1954).

[2] King, E. J., and Wootton, I. D. P., "Micro-analysis in Medical Biochemistry", third ed. (Churchill, 1956).

Chromatography of Xanthene Dyes

RECENTLY, Lima and Pieroni[1] have directed attention to the heterogeneous character of the dye rose bengal, which, when labelled with iodine-131, is used in tests of hepatic function[2,3]. Applying the ethanol–ammonia solvent system of Ishida *et al.*[4], they claim resolution of this dye into four components, three coloured and one colourless.

In the course of an investigation of the phloxin/tartrazine method, described by Lendrum[5] for the staining of connective tissues, I have had occasion to examine chromatographically most of the xanthene dyes in common use, and feel that my results may be of interest in relation to the observations quoted above.

A slight modification of the solvent system of Ishida *et al.*[4] was found to be the most useful ; this consisted of 0·880 ammonia, ethanol and water in the proportion 5 : 10 : 85 (by volume). Paper chromatograms were run on Whatman No. 1 paper 20 × 40 cm. by the descending method of Consden *et al.*[6] with the modification that the dye was applied as a thin line at the starting point. Circular chromatograms by a variation[7] of the method of Rutter[8] were found to be convenient for the rapid comparison of different batches and gave good separations. R_F values tended to show some variation, and the values quoted below are the mean values of several results.

Commercial halogenated fluoresceins examined included eosin *Y* ([*R*]-tetrabromofluorescein), erythrosin *R* ([*R*]-di-iodofluorescein) and erythrosin *B* ([*R*]-tetraiodofluorescein). The first of these dyes showed two coloured components, R_F values 0·16 and 0·22 ; the second gave five components, R_F values 0·08, 0·15, 0·26, 0·49 and 0·64 ; the third dye gave two components with R_F values 0·08 and 0·15.

Studies made on the partial halogenation of fluorescein with bromine and iodine in aqueous alkali showed that even at the stage where only one molecular proportion of halogen had been added, the reaction mixture could be resolved chromatographically into five coloured components, the position of the absorption maxima of which decreased *pari passu* with increase of R_F value. The R_F values of these components were, after bromination: 0·16, 0·22, 0·33, 0·52 and 0·64; after iodination: 0·08, 0·15, 0·26, 0·49 and 0·64. The components with R_F 0·64 were identical and could not be distinguished on chromatographic or spectroscopic criteria from fluorescein. It would seem a reasonable assumption that the four other components in each case represent the mono-, di-, tri- and tetra-halogenated fluoresceins. Further halogenation of the initial mixture resulted in a gradual decrease in the proportion of components of high R_F value. The end-product in each case had two coloured components which appeared to be identical with those of commercial samples of eosin *Y* and erythrosin *B*, respectively.

Essentially similar results were obtained in experimental halogenation of [*P*]-tetrachlorofluorescein. The R_F values for the components were in this case: bromination: 0·40, 0 48, 0·59, 0·71 and 0·77 ([*P*]-tetrachlorofluorescein); iodination: 0·31, 0·42, 0·54, 0·71 and 0·77. A commercial sample of rose bengal (extra) had two coloured components with R_F values of 0 31 and 0·42; these were presumably the tri- and tetra-iodo[*R*]-tetrachloro[*P*]-fluoresceins. Six samples of commercial phloxin *B* (*C.I.* No. 778) were examined. These were generally more complex mixtures than the dyes mentioned above. Some of the fractions could be identified with the products of bromination of [*P*]-tetrachlorofluorescein and in one case there appeared to be admixture with iodinated fluoresceins. It is possible that the other components are derivatives of [*P*]-dichlorofluorescein, which is stated[9] to be the basis of phloxin (*C.I.* No. 774).

In addition to the multiplicity of coloured components in these dyes, an examination of the chromatograms in ultra-violet light revealed the presence of a number of colourless components which showed brilliant fluorescence. This was especially marked in the case of derivatives of [*P*]-halogenated fluorescein. In some of the samples of phloxin *B*, as many as four such components could be found.

In view of the statement by Stowe, Delprat and Weeks[10] that the liver only eliminates rose bengal when this latter is fully halogenated, a careful chromatographic examination would appear to be essential in this and similar applications of the xanthene dyes.

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[1] Lima, F. W., and Pieroni, R. R., *Nature*, 184, 1065 (1959).

[2] Blahd, W. H., and Nordyke, R. A., *Clin. Res. Proc.*, 5, 40 (1957).

[3] Taplin, G. V., Meredith, O. M., and Kade, H., *J. Lab. Clin. Med.*, 45, 665 (1955).

[4] Ishida, M., Inagaki, S., and Watanabe, E, *J. Pharm. Soc. Japan*, 73, 736 (1953).

[5] Lendrum, A. C., *J. Path. Bact.*, 59, 390 (1947).

[6] Consden, R., Gordon, A. H., and Martin, A. J. P., *Biochem. J.*, 38, 224 (1944).

[7] Taylor, K. B., *J. Histochem. Cytochem.* (in the press).

[8] Rutter, L., *Nature*, 163, 487 (1949); *Analyst*, 75, 37 (1950).

[9] Conn, H. J., "Biological Stains" (Biotech. Publications, Geneva, N.Y., 1953).

[10] Stowe, W. P., Delprat, G. D., and Weeks, *Amer. J. Clin. Path.*, 3, 55 (1933).

PHYSIOLOGY

Hormonal Control of Moulting in Amphibians

MOULTING in amphibians starts when a new stratum corneum of the epidermis is formed. When moulting is normal the old stratum corneum is loosened and the slough thus formed is finally shed. (In the literature on amphibians, 'moulting' is often used synonymously with 'sloughing', that is, to denote merely the shedding of the slough.)

It is well known that moulting is abnormal in hypophysectomized amphibians. This is especially conspicuous in toads and most urodeles where hypophysectomy prevents shedding of the slough, whereas formation of strata cornea continues; eventually a dark-brown, thick, cornified layer covers the skin.

In urodeles thyroidectomy inhibits sloughing as effectively as does hypophysectomy. Reversely, sloughing can be produced by injection of thyrotropic hormone into hypophysectomized animals or of thyroxin into hypophysectomized or thyroidectomized animals[1–3]. Sloughing was also caused by thyroxin in hypophysectomized *Bufo arenarum*, but was not inhibited by thyroidectomy[4]. Sloughing was not inhibited by thyroidectomy or treatment with thiourea or thiouracil in *B. regularis* (Ewer, D. W., personal communication), or by treatment with thiourea or thiocyanate in *B. bufo* (Rosenkilde, Per, personal communication).

During investigations on hypophysial function in the toad *B. bufo* we observed that injection of adrenocorticotrophic hormone could cause sloughing in hypophysectomized animals. It was therefore found of interest to compare the effects of thyroxin and adrenocorticosteroids on moulting after hypophysectomy in a urodele (*Amblystoma mexicanum* Cope) and in a toad (*B. bufo* (L)).

Normally, *A. mexicanum* does not metamorphose. The skin of the unmetamorphosed animals does not hyperkeratinize in response to hypophysectomy in contrast to the skin of metamorphosed animals[3]. Metamorphosis was therefore induced by injection of 50 μgm. thyroxin. The animals of both species weighed around 50 gm. In *Bufo* only pars distalis of the hypophysis was extirpated; in *Amblystoma* the neuro-intermediate lobe was generally removed together with the pars distalis, because the two parts are not easily separated in this species. The technique of operation in *Bufo* has been described previously[5]. A similar technique was used in *Amblystoma*. Injections were given subcutaneously of thyroxin (Roche) and "Decorton" (Organon, deoxycorticosterone acetate, D.C.A., dissolved in peanut oil). Control injections of peanut oil were made.

In hypophysectomized *Amblystoma*, 50 μgm. thyroxin caused sloughing 2–3 days after injection, whereas in hypophysectomized *Bufo* 50 μgm. or 100 μgm. thyroxin had no effect (Table 1). In contrast, deoxycorticosterone acetate caused sloughing in *Bufo*, but not in *Amblystoma*. Both in *Amblystoma* and *Bufo* 0·5 mgm. deoxycorticosterone acetate was given twice a week for several weeks. The slough was not shed in *Bufo* after every injection, but when sloughing occurred it was always within 24 hr. after the injection. In seven toads 33 moults were produced; one toad died a week after hypophysectomy without sloughing. Control injections of oil had no effect on the skin. Mostly the sloughs were eaten

Table 1. EFFECT OF THYROXIN AND DEOXYCORTICOSTERONE ACETATE ON SHEDDING OF CORNIFIED LAYER OF EPIDERMIS IN HYPOPHYSECTOMIZED *Bufo bufo* AND *Amblystoma mexicanum*

Treatment	Species	Dose (mgm.)	No. of animals in which cornified layer was Shed	Not shed
Thyroxin	*B. bufo*	0.05	0	8
		0.1	0	6
	A. mexicanum	0.05	5	0
Deoxycorticosterone acetate	*B. bufo*	0.5	7	1
	A. mexicanum	0.5	0	5

as they are during normal moulting. However, the injections did not restore the normal moulting rhythm. New strata cornea were produced two or more times a week as in untreated hypophysectomized toads, whereas the normal controls moulted only about once a week.

It is probably not justified to conclude from the present experiments that sloughing is dependent exclusively on the thyroids in *Amblystoma* and on the adrenals in *Bufo*. Both glands may play a part in sloughing as well as other phases of moulting in both *Amblystoma* and *Bufo*, though seemingly to varying degrees.

In the literature it is more often tacitly understood than directly expressed that the moulting cycles in amphibians are maintained and controlled by rhythmic activity of the thyroids. Adams *et al.*[6], however, clearly state that they find it likely that the moulting rhythm in *Triturus* "is dependent at least in part on an endocrine (pituitary–thyroid) balance". Scharrer[7] considers the moulting rhythm in *Bufo* to be regulated by a special centre in the central nervous system, the centre acting via the hypophysis and the thyroids.

However, the following observations do not support the assumption that the single moult is normally initiated by increased activity of the thyroids—or other endocrine glands.

(1) In metamorphosed *Amblystoma* the transplanted hypophysis is able to maintain the thyroid activity at a level that permitted the normal moulting cycles to continue. The glands were transplanted under the palate mucosa and could therefore scarcely be rhythmically stimulated by any moulting centre[8].

(2) Injection of thyroxin into normal *Amblystoma* or *Bufo* did not immediately cause sloughing or loosening of the stratum corneum, but the moulting frequency was raised during the week or weeks following a single injection. This was especially pronounced in *Amblystoma*, which moulted almost daily for about a week—or some five times faster than normally—after a single injection of 50 μgm. thyroxin (Table 2). After about four moults in rapid succession sloughing stopped completely for up to three weeks. During this period the epidermis became covered by a thick cornified layer which was finally shed spontaneously; and an almost normal moulting rhythm was resumed. The observations are naturally explained by assuming that the high level of thyroxin in the blood immediately after the injection enhanced the rate of moulting, but at the same time inhibited the activity of the thyroids for several days so that the level of endogenous thyroid hormone did not permit sloughing. Shedding of the sloughs continued as long as sufficient exogenous thyroxin remained in the blood, and after this period did not start until the animal's own thyroids functioned again. In the hypophysectomized *Amblystoma* the single injection of thyroxin also initiated a burst of moults.

Table 2. NUMBER OF MOULTS PER WEEK IN FIVE *Amblystoma mexicanum*

	Before	After injection of 50 μgm. thyroxin (weeks)						
		1	2	3	4	5	6	7
Normal animals	6*	18	2	0	3	7	8	8
Hypophysectomized animals	0	21	0	0				

*Average of the seven weeks that preceded the injection.

(3) Finally, it should be stressed that hypophysectomy inhibits the shedding, but not the formation of the slough; new strata cornea are still being formed and even faster than normally.

Thus normal moulting depends upon or may be influenced by the function of the thyroids, adrenal cortex and perhaps other endocrine glands as well[9]. The frequency of moulting is determined by the level of activity of the thyroids and not by a rhythm of activity; the ability to moult rhythmically is apparently inherent in the skin.

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[1] Adams, A. E., and Gray, B., *Anat. Rec.*, 65, 69 (1936).

[2] Osborn, C. M., *Anat. Rec.*, 66, 257 (1936).

[3] Taylor, A., *J. Exp. Zool.*, 73, 153 (1936).

[4] Ungar, I., *C.R. Soc. Biol.*, 112, 504 (1933).

[5] Jacobsohn, D., and Jørgensen, C. B., *Acta physiol. Scand.*, 36, 1 (1956).

[6] Adams, A. E., Kunder, A., and Richards, L., *J. Exp. Zool.*, 63, 1 (1932).

[7] Scharrer, E., in "Comparative Endocrinology", ed. by Gorbman, A., 233 (John Wiley and Sons, Inc., New York, 1959).

[8] Jørgensen, C. B., Larsen, L. O., Rosenkilde, P., and Wingstrand, K. G. (unpublished work).

[9] Chadwick, C. S., and Jackson, jun., H. R., *Anat. Rec.*, 101, 718 (1948).

Selective Biological Membrane Transport of Individual Amino-Acids

THE behaviour of amino-acids in high-tension (100–300 V./cm.) electrostatic fields has recently been investigated in this laboratory. A phenomenon (to be described shortly) was observed, and it led to a hypothesis which is summarized in the following.

Since the Donnan effect cannot explain the selectivity of membrane transport of molecules larger than mono- or di-atomic ions and the Michaelis theory[1] becomes more and more[2] justified, we think our observations have given us an explanation of many problems, among them, for example, why cells put into a concentrated solution of amino-acids take up only a determined amount of amino-acid and not more[3]. How can it be explained by the Donnan effect why cells do take up only specified amino-acids or other molecules, and why can one or the other be substituted by a completely different kind of molecule[4], etc.?

However, to-day we know more about the structure of some biologically important membranes. The electron microscope has shown that phospholipid molecules are arranged[5] in the anterior pituitary gland cell wall in a laminary way, 20 Å. apart from each other in triplets, making up a thickness of 75 Å. Halpern and Binaghi have shown a selective permeability of certain biological membranes[3] to carbonic acid, and a similarly selective activity of the human erythrocyte membrane towards uric acid has been demonstrated[6], and further that calcium ion has an important role in maintaining this selective permeability was shown by Maizels[7]. Putting together the findings of the above workers and several others as well as ours we came to the following conclusion.

An amino-acid—let us keep for the sake of simplicity to this example—which has been liberated from the food protein by digestive ferments finally acquires an electrostatic charge in the jejunum. The amino-acid will then approach the wall of the gut (and it must not get nearer to it than 50 Å.[8]) at a point where there is a venous capillary or lymph duct on the other side. At these points the inside of the gut membrane will have a polarity and potential sufficiently great enough to attract the amino-acid, which bears an opposite polarity and has enough reserve energy[9] to alter its structure into another form—an elongated one most probably. This enables it to pass now the approximately 20-Å. wide opening on the membrane towards which it is pulled. As soon as the amino-acid reaches the other side of the membrane it will be pushed away since this side has the same polarity as the amino-acid, and will travel along by now, let us say, in the venous capillary, returning to its original structure. Since the *p*H value of the blood will differ from that in the gut, the amino-acid receives a new charge and also, possibly, alters its polarity.

The electrostatic structure of the ribo-nucleus molecule in a given cell, at some distance from our amino-acid, will cause the production by that cell of a certain quadruple peptide. When three given amino-acids in this cell are already linked together, a certain pattern of polarity and electrostatic charge will be set up, which will affect the cell membrane either as a whole or only a part of it. It is necessary for this hypothesis that this individual cell membrane should be some ten times more sensitive to changes in electrostatic charge than the gut membrane. This will enable the cell to discriminate between individual amino-acids. When the 'missing' amino-acid reaches the vicinity of the cell it will be attracted to it and will enter into it with the same type of changes as were described when it passed through the gut membrane, except that when it now reaches the cell interior, instead of moving around freely it will immediately combine with the already coupled group of amino-acids, thereby changing the electrostatic state of the cell again. The completed quadruple peptide will then be either incorporated structurally into the cell, or the polar and electrostatic changes will enable the peptide to pass through the cell membrane and get into the blood circulation, where it can be used up for any further macro-molecular building.

This selectivity of membrane transport for amino-acids has been demonstrated in this laboratory on a small-scale, simple model-experiment. Two amino-acids, histidine and aspartic acid, were dissolved in a buffer solution at a *p*H of 8·6 and kept at 38° C. with a constant slow stirring. Into it, five tubes were dipped, the lower end of each being covered by a 'Cellophane' membrane. The tubes contained the same buffer solution but the *p*H was varied in steps of 0·2 from 7·0 to 7·8. At the end of 5 hr. it was found that all aspartic acid was in the tube of *p*H 7·0 and all the histidine in the tube of *p*H 7·2. No amino-acid was detected in any of the other three tubes, or in the original buffer solution.

Further work on this problem is being carried out with the close co-operation of the Marconi Instruments Co., Ltd., on a larger and more critical scale. It is hoped that this further stage of the model-experiment will throw more light on the problem of specific membrane transport.

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[1] Michaelis, M., and Fujita, A., *Biochem. Z.*, **161**, 47 (1925).
[2] Butkewitch, W. L. S., *Pflugers Arch.*, **161**, 468 (1925).
[3] Halpern, B., and Binaghi, R., *Nature*, **183**, 1397 (1959).
[4] Christensen, H. N., *et al.*, *J. Biol. Chem.*, 198, 1, 17 (1952).
[5] Barnes, G. B., *Nature*, **184**, 651 (1959).
[6] Overgaard-Hansen, K., and Lassen, U. V., *Nature*, **184**, 554 (1959).
[7] Maizels, M., *Nature*, **184**, 366 (1959).
[8] Potter, G., and Weber, C., *Nature*, **184**, 688 (1959).
[9] Schellman, J. A., *C.R. Lab. Carlsberg, Sér. Chim* , 29, 229 (1956).

Induction of a Vascular Permeability Factor by the Interaction of Guinea Pig Tissue Debris with Serum

THE sera of the guinea pig[1,2], rat[3], rabbit[3] and man[4] contain the inactive precursor (*pro-PF*) of a globulin permeability factor (*PF*) activable by dilution in saline (*PF/Dil*) or by chemical fractionation. The *in vivo* mechanism of activation is unknown; but an obvious hypothesis—that activation in injury is induced by contact of the *pro-PF* with damaged tissue—was investigated by testing the effect on guinea pig serum of debris from homogenates of guinea pig tissues.

Guinea pig serum was harvested from defibrinated blood[5] and stored at 2° C. The excised tissues were freed of adherent connective tissue bearing blood vessels and disrupted[6] in sterile 0·85 per cent saline, using a loosely fitting homogenizer of the Potter type. The homogenate was centrifuged at 3,000*g* for 20 min. and the supernatant decanted. The residual tissue debris, twice washed in 20 vol. saline, was suspended in saline, such that 1 gm. wet tissue yielded 1·5 ml. tissue debris suspension. Dried tissue debris was obtained by adding the suspension to 10 vol. acetone, removing the debris 10 min. later by filtration and drying it in a stream of air. Permeability-increasing activity of the preparations was estimated in terms of the diameter of the area of exudation of circulating pontamine blue induced at the site of intracutaneous injection in depilated guinea pigs[7].

The ready and progressive activation of *pro-PF* to *PF/Dil* by dilution of normal serum in saline depends on the age and temperature of the dilutions[1]. In preparations held 90 min. at room temperature, maximum permeability-increasing activity occurs in 1/150 serum, but at this time low dilutions of serum, 1/1–1/15, have no substantial activity because activated *PF* has been antagonized by a low-potency, slowly acting inhibitor (*IPF*) present therein. A second factor (*PF/Age*)[1] matures in serum kept for one or more days. *PF/Dil* is characterized by its insusceptibility to the antihistamine, mepyramine maleate, and its high susceptibility to trypsin inhibitors like that from soya bean (*SBTI*); *PF/Age* is not antagonized by *IPF*, *SBTI* or mepyramine.

Tissue debris suspensions of lung, liver, heart, skin, brain and skeletal muscle each were mixed with 2·5 vol. serum and after 15 min. the debris was removed by centrifugation. All except skeletal muscle induced permeability-increasing activity in the treated serum (Table 1). The most active, brain debris, was investi-

Table 1. PERMEABILITY-INCREASING ACTIVITY OF SERUM TREATED WITH HOMOGENATES OF VARIOUS TISSUES

Tissue homogenate	Mean diameter (mm.) of area of increased permeability induced by: Saline extract	Treated serum (1/3)
Lung	2 9	5·5
Liver	2·6	5·6
Heart	2·8	6·1
Skin	2 9	6 6
Brain	3 5	7·8
Skeletal muscle	3·9	3 9
None	—	4·2

gated. Mixed with normal serum, within 5 min. it led to the appearance of a new permeability factor in the 1/1–1/15 dilutions. The amount of factor produced was proportional to the amount of brain debris added; and 1 hr. exposure to debris was no more effective than 5 min. The new factor produced by the reaction of debris with serum differed from *PF/Dil* in being insusceptible to *SBTI*, only slightly susceptible to serum fractions containing *IPF*, but highly susceptible to mepyramine. In fact, the addition of mepyramine to dilutions of treated serum restored the pattern of activity to that of untreated serum. These results also distinguish the new permeability factor from *PF/Age*.

Saline extracts of dried brain debris and the supernatant fluid from brain homogenate also contain a permeability factor with the same susceptibility to inhibitors. Moreover, all three preparations—fresh debris mixed with serum, the supernatant from fresh debris and a saline extract of dried debris—induced a slow contractor response in atropinized guinea pig ileum *in vitro*, which was not suppressed by mepyramine; and on injection into the rat peritoneal cavity[8], all three readily released histamine (Table 2). That is, the new factor appears to be a histamine-liberator. Prepared from saline extracts of dried debris, it is stable for 10 min. at 60°, 80° or 100° C. It does not appear to be associated with a protease system (for example, plasmin) because *SBTI*, 333 μgm./ml., also fails to inhibit its appearance in mixtures of serum and debris.

Table 2. HISTAMINE RELEASED IN THE RAT BY THE INTRAPERITONEAL INJECTION OF DEBRIS FACTOR AND OTHER PREPARATIONS

Substance injected	Histamine yield Exudate (μgm./ml.)	Body-weight (μgm./100 gm.)
Compound 48/80 (ref. 11) 15 μgm./ml.	0 20	1 03
Debris in 1/3 serum	0 24	0·94
Untreated 1/3 serum	0 00	0·00
Supernatant of brain homogenate*	0·15	1·00
Dried debris	0·13	0 42
Locke's solution	0·03	0·14

* Prepared in Locke's solution.

It is suggested that many tissues normally contain a bound histamine-liberator, which is released on mechanical disruption, after treatment with acetone or by the action of serum on damaged tissue. Moreover, the results provide a model for a tissue mechanism of histamine liberation—serum releasing histamine directly from injured tissue, or activating the factor, which in turn liberates histamine. *In vitro* the reactions are rapid; release of the factor by serum is complete in 5 min., the shortest period tested; and the released factor, injected intracutaneously, increases permeability in ½ min.

The new factor may mediate the immediate permeability response of tissues to injury[9]. However, it is not possible at this stage to evaluate the relation of this system to the superficially similar one described by Spector[10] in the serum and pulmonary mitochondria of the rat, and ascribed by him to an activation of serum *pro-PF* by mitochondria.

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[1] Miles, A. A., and Wilhelm, D. L., *Brit. J. Exp. Path.*, 36, 71 (1955).
[2] Wilhelm, D. L., Miles, A. A., and Mackay, Margaret E., *Brit. J. Exp. Path.*, 36, 82 (1955).
[3] Wilhelm, D. L., Mill, P. J., Sparrow, Elizabeth M., Mackay, Margaret E., and Miles, A. A., *Brit. J. Exp Path.*, 39, 228 (1958).
[4] Mill, P. J., Elder, J. M., Miles, A A., and Wilhelm, D. L., *Brit. J. Exp. Path.*, 39, 343 (1958)
[5] Wilhelm, D L, Mill, P. J., and Miles, A. A., *Brit. J. Exp. Path.*, 38, 446 (1957).
[6] Darbre, A, *J. Physiol.*, 136, 2*P* (1957).
[7] Miles, A. A., and Miles, E. M., *J. Physiol.*, 118, 228 (1952).
[8] Fawcett, D. W., *J. Exp. Med.*, 100, 217 (1954).
[9] Wilhelm, D. L., and Mason, Brenda, *Brit. Med. J.*, ii, 1141 (1958).
[10] Spector, W. G., *J. Path. Bact.*, 74, 57 (1957).
[11] Paton, W. D. M., *Brit. J. Pharmacol.*, 6, 499 (1951).

Ataxia in Hens poisoned by Tri*para*ethylphenyl Phosphate

IT has been known for almost 30 years that the administration of tri*ortho*cresyl phosphate and certain other organo-phosphorus compounds produces demyelination in the central and peripheral nervous system of some species. Earl and Thompson[1] suggested that the lesions might be associated with the anticholinesterase activity of the drugs *in vivo*, but Davison[2] later demonstrated that some other organo-phosphorus compounds, which were equally effective as inhibitors of cholinesterase, did not cause demyelination or ataxia. More recently another arylphosphate, tri*para*ethylphenyl phosphate, has been synthesized, which causes ataxia in hens[3], but, unlike the other demyelinating agents of this class, it does not possess demonstrable anticholinesterase activity (Aldridge, W. N., and Barnes, J. M., personal communication). The results with this material appear to provide conclusive evidence that the organo-phosphorus compounds do not produce demyelination by virtue of their anticholinesterase effects.

However, it may be wrong to assume that the toxic action of all the demyelinating drugs in this group is exactly the same. In this laboratory, it has been observed that the clinical picture produced in hens dosed with tri*para*ethylphenyl phosphate differs very strikingly from that seen with tri*ortho*cresyl phosphate poisoning. In hens poisoned by tri*ortho*cresyl phosphate, ataxia is associated with very marked flexion of the joints of the leg and with curling of the toes. In contrast to this, extensor tone is greatly increased in the hens dosed with tri*para*ethylphenyl phosphate; these stand on tiptoe and walk with exaggerated extension of the legs. In the late stages, when their legs are unable to support their weight, the birds sit with their legs extended rather than flexed. Another difference is in the time course of the condition; in tri*ortho*cresyl phosphate poisoning changes in gait are never seen before the seventh day after dosing, but once symptoms appear the condition progresses rapidly and many birds became moribund during the third or fourth week. Only those birds with relatively mild symptoms survive, and recovery is extremely slow; for example, one bird was still ataxic 27 weeks after dosing. On the other hand, symptoms appear as early as the fourth day after tri*para*ethylphenyl phosphate has been given but they increase more slowly and less steadily than in tri*ortho*cresyl phosphate poisoning. While some of the birds with severe disabilities have died, others have shown obvious improvement from the fourth to fifth week and in some less-affected hens recovery has been almost complete 7 weeks after dosing.

Although no claim is made on the basis of this work that tri*para*ethylphenyl and tri*ortho*cresyl phosphates produce their effects by a different biochemical pathway, the findings suggest that such a possibility should be considered. In view of this it seems important that workers in this field should describe their clinical observations more precisely than hitherto. It may be wrong to classify all organo-phosphorus-produced disabilities as 'Ginger Jake paralysis'.

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[1] Earl, C. J., and Thompson, R. H. S., *Brit. J. Pharmacol.*, 7, 261 (1952).

[2] Davison, A. N., *Brit. J. Pharmacol.*, 8, 212 (1953).

[3] Bondy, H. F., Field, E. J., Worden, A. N., and Hughes, J. P. W. *Brit. J. Indust. Med.* (in the press).

Glucagon : a Protein Catabolic Hormone in the Isolated Perfused Rat Liver

WE have reported that the isolated perfused rat liver from a fasted animal produces urea in linear fashion[1], at the expense of both liver and plasma proteins. Urea production occurs, and the blood amino-acid level is maintained in the absence of added extraneous amino-acids. When a free amino-acid supplement is added, urea production is greatly enhanced. The addition of glucose or fructose supplements is associated with a nitrogen-sparing effect manifest as a diminished production of urea. This nitrogen-sparing effect is seen in either the presence or absence of added amino-acids.

The production of urea by the isolated perfused liver may be regarded as a measure of the continuing or endogenous protein catabolism, and the isolated perfused liver affords a means of studying protein catabolism and the effects of various agents upon it.

This communication reports a remarkable effect of the hormone glucagon (generously supplied by the Eli Lilly Co.) in enhancing protein catabolism in the isolated perfused liver.

In typical experiments a 6-hr. liver perfusion is started and continued for a base-line period of 1½–2 hr. During this time, Ringer's solution is continuously infused. After this the continuous infusion is changed to one of Ringer's solution containing glucagon in such concentration that as little as 1 μgm. of glucagon is infused per hour.

Blood samples are removed at regular intervals, analysed for urea by Conway's method and for glucose by Nelson's method. The total amount of urea produced during each interval is computed after appropriate correction for the amount of urea removed by the sampling procedure itself.

Not only is prompt glycogenolysis produced by as little as 1 μgm. of glucagon per hour, but also, as Fig. 1 shows, the rate of urea production is greatly increased. Almost complete glycogenolysis occurs in the face of blood glucose-levels known to promote glycogen deposition[2] and also to spare protein[1].

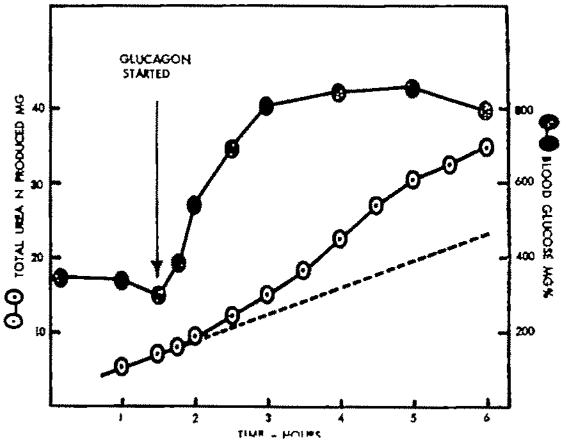

Fig. 1. Liver donor fed stock laboratory diet. Blood donors fasted 18 hr. before being bled to provide a total of 63 ml. of whole blood diluted with 35 mgm. heparin and Ringer's solution to 79 ml. at start of perfusion. Perfused liver weight, 14 gm.; glycogen content at 1½ hr. 2·64 gm. per cent, and at 6 hr. 0·11 gm. per cent. Over an interval of 1½–6 hr. glucagon was infused at the rate of 1 μgm. per hr. in 3 ml. of Ringer's solution

It is of some interest that the hormone hydrocortisone in the form of the soluble succinate given by continuous infusion in amounts up to 2 mgm. per hour produces no change in the output of urea by livers from either normal or adrenalectomized rats. Only when glucagon is infused into livers from the latter along with hydrocortisone is a maximal increase in protein catabolism elicited. Given alone, in perfusion of the adrenalectomized rat liver, glucagon causes a small but definite increase in protein catabolism.

The glycogenolytic effect of glucagon at 1 μgm. per hour on the liver is not altered by prior and continuous administration of insulin in amounts known to be sufficient to enhance glucose uptake and lipogenesis[3]. However, the protein catabolic effect of 1 μgm. per hour is abolished by the insulin.

Although relatively massive doses of glucagon given parenterally to intact rats have been associated with enhanced urinary nitrogen excretion[4], the site of action and the mechanism of the response have not been defined. Our experiments implicate the liver directly and point to glucagon as being a protein catabolic hormone *par excellence*, with adrenal corticoid hormone exerting a permissive action on this effect.

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[1] Miller, L. L., Burke, W. T., and Haft, D. E., *Fed. Proc.*, 14, 707 (1955).

[2] Sokal, J. E., Miller, L. L., and Sarcione, E. J., *Amer. J. Physiol.*, 195, 295 (1958).

[3] Haft, D. E., and Miller, L. L., *Amer. J. Physiol.*, 192, 33 (1958).

[4] Salter, J. M., Davidson, I. W. F., and Best, C. H., *Diabetes*, 6, 248 (1957).

Effects of Reserpine, Serotonin and Vasopressin on the Survival of Cold-Stressed Rats

In the preparatory stages of a projected study of the hypothalamic control of release of adrenocorticotrophic hormone, large doses of reserpine, serotonin creatinine sulphate, and commercial vasopressin were administered to rats which were then subjected to cold stress. Preliminary findings are reported here in view of the widespread interest in the drugs from the point of view of pituitary-adrenal physiology[1] and in view of the current extensive use of reserpine in medicine.

Intact male rats of the Harvard strain weighing 95–105 gm. were selected. All substances were administered subcutaneously, and cold-exposed controls received volumes of vehicle equal to those injected into the test animals. The rats were placed in individual screen-bottomed cages without food or water in a cold room at 3° ± 1° C. either immediately following the injection, or in some cases 18 hr. after injection. To minimize variations due to cyclic changes in physiological activity the animals were placed in the cold room at approximately the same time (1200 midnight ± ½ hr.) except for one stressed control group which was exposed to cold starting at 11 a.m. Treated controls were kept at room temperature under otherwise similar conditions. For the sake of comparison a group of adrenalectomized animals (1 week after surgery) in the same weight range was similarly stressed. The results are summarized in Table 1.

It is evident that large doses of reserpine, serotonin creatinine sulphate, and commercial vasopressin effectively lower the ability of intact rats to withstand cold stress, although on the basis of the experiments described here it is not possible to offer any demonstrable explanation for the mechanisms involved; however, the similarity in the survival-times of serotonin- and vasopressin-treated rats and the survival times of adrenalectomized animals suggests that treatment with these two drugs—both of which are known to exert some action on the pituitary-adrenal system[1]—results in temporary functional adrenalectomy.

Table 1. EFFECTS OF RESERPINE, SEROTONIN CREATININE SULPHATE AND VASOPRESSIN ON THE SURVIVAL-TIMES OF COLD-STRESSED RATS

Treatment	Survival time*	No. of animals
1 Intact controls, exposed at 1200 midnight	19·25 ± 1·8 hr.	20
2 Intact controls, exposed at 11 a.m.	9 56 ± 1·0 hr.	9
3 Adrenalectomized, untreated animals	2·98 ± 0 8 hr	9
4 2 mgm. serotonin creatinine sulphate, stressed immediately after injection	< 4 hr.	10
5 1 mgm. serotonin creatine sulphate, stressed immediately after injection	< 4 hr.	10
6 1 mgm. serotonin creatinine sulphate, stressed immediately after injection	3 57 ± 0·6 hr.	7
7 2 mgm, serotonin creatinine sulphate, stressed 18 hr. after injection	killed after 15 hr.	8
8 1 mgm. serotonin creatinine sulphate, stressed 18 hr. after injection	killed after 15 hr.	8
9 1 mgm. reserpine, stressed immediately after injection	9·5 ± 0·6 hr.	18
10 1 mgm. reserpine, stressed 18 hr. after injection	10·5 ± 0 7 hr.	10
11 3 units vasopressin, stressed immediately after injection	2·08 ± 0·15 hr.	6
12 2 units vasopressin, stressed immediately after injection	3·06 ± 0·53 hr.	9
13 3 units vasopressin, stressed 18 hr. after injection	killed after 15 hr.	6
14 2 units vasopressin, stressed 18 hr. after injection	killed after 15 hr.	6

All treated controls kept at room temperature survived for 24 hr. and were killed at that time.

* Mean ± standard error of the mean.

Reserpine clearly differs from the other substances tested in that its effects are still evident 18 hr. after injection, although most of the drug is known to have been metabolized by then[2]. The survival times of intact controls exposed to cold stress at different times of the day appear to reflect the daily cyclic changes in adrenal activity[3].

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[1] Shibusawa, K., Saito, S., Fukuda, N., Kawai, T., and Yoshimura, F., *Endocrinol. Jap.*, 2, 47 (1955). Wells, H., Briggs, F. N., and Munson, P. L., *Endocrinol.*, 60, 571 (1957). Harwood, C. T., and Mason, J., *ibid.*, 60, 239 (1957).

[2] Numerof, P., Gordon, M., and Kelly, J. M., *J. Pharmacol. and Exp. Therap.*, 115, 427 (1955).

[3] Vermund, H., Halberg, F., Barnum, C. P., Nash, C. W., and Bittner, J. J., *Amer. J. Physiol.*, 186, 444 (1956). Albrecht, P., Visscher, M. B., Bittner, J. J., and Halberg, F., *Proc. Soc. Exp. Biol. and Med.*, 92, 703 (1956).

Potency of Parenteral Vitamin A

Vitamin A is widely employed in clinical practice and is also used extensively as a teratogenic agent for the production and study of fœtal abnormalities in laboratory animals[1].

Up to the present the customary procedure in experimental work has been for the vitamin to be dissolved in oil and administered orally. This method has a number of disadvantages. In order to ensure that the animal receives the required dose, it is necessary to put the dissolved vitamin directly into the stomach by the somewhat laborious procedure of gastric intubation. Once in the alimentary tract, defects of absorption may lead to the loss of a substantial amount of the vitamin in the fæces. Such defective absorption may be due to the presence of gastric or intestinal inflammatory conditions, to a deficiency in the secretion of bile which leads to a failure in the breakdown of the fat in which the vitamin is dissolved, or to the nature of the oil itself[2]. Some of these factors may also militate against absorption of the vitamin when employed for therapeutic purposes in man.

Subcutaneous or intramuscular injection has many advantages in experimental work from the point of view of convenience and the more precise measurement of dosage, but the results of the injection of oily solutions of vitamin A have been disappointing. Brown and Sturtevant[3] found that during a 28-day test period little if any benefit resulted from injection and concluded that vitamin A in oil injected subcutaneously or intramuscularly was utilized to a very small extent if at all. Their conclusions are borne out by the results of experiments which we have carried out in rats. The subcutaneous injection into rats of 60,000 I.U. vitamin A dissolved in arachis oil daily from the eighth to the thirteenth day inclusive of pregnancy did not produce any cleft palates in the young, whereas the same quantities given by gastric intubation over the same period of time resulted in 29·7 per cent of cleft palates in the young[4].

Recently we have had the opportunity of studying the teratogenic effects of the subcutaneous injection of vitamin A stabilized in 'Tween 80'. Female rats of the Wistar strain were used. After mating with normal male rats, pregnancy was determined by the presence of spermatozoa in the vagina. The pregnant females received 60,000 I.U. vitamin A (suspended in 'Tween 80') daily by subcutaneous injection on the twelfth, thirteenth and fourteenth days of pregnancy.

The rats were killed on the twentieth day of pregnancy and the fœtuses removed from the uteri for examination. The results of the two pilot experiments in relation to malformations of the palate are shown in Table 1. In addition to the palatal deficiencies many of the young showed other skeletal malformations involving mainly reduction of the digits and syndactly.

Table 1 INCIDENCE OF CLEFT PALATE FOLLOWING PARENTERAL VITAMIN A

Group	No. of dams	Total No. of young	No of young with cleft palates	Percentage of young with cleft palates
1	8	68	49	72·0
2	7	54	49	90·7

The percentage incidence, in the fœtuses, of cleft palates produced by the subcutaneous injection of vitamin A compares closely with that of 92 per cent observed by Giroud and Martinet[5] when female rats were given 60,000 I.U. vitamin A, in oily solution, by mouth on the eleventh, twelfth and thirteenth days of pregnancy. The disparity in the percentage incidence of the malformation in our two groups of animals is probably due to the small numbers used in these pilot experiments, although some other unknown factor may be involved.

These experiments indicate that vitamin A suspended in 'Tween 80' is potent as a teratogen when injected subcutaneously. From the experimental point of view, the advantages of this route of administration over the oral are obvious. It may also be useful, at times, to be able to employ a parenteral route for therapeutic purposes.

This work has been supported by a grant from the Medical Research Council, which is gratefully acknowledged. We wish to thank Messrs. Vitamins, Ltd., for the preparation and gift of the vitamin solution.

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[1] Cohlan, S. Q., *Science*, 117, 535 (1953).

[2] Bicknell, F., and Prescott, F., "The Vitamins in Medicine" (Heinemann, 1947).

[3] Brown, R. A., and Sturtevant, M., *Vitamins and Hormones*, 7, 171 (1949).

[4] Woollam, D. H. M., and Millen, J. W., *Brit. Med. J.*, ii, 197 (1957).

[5] Giroud, A., and Martinet, M., *Arch. Anat. Micro.*, 45, 77 (1956).

The Nuclear Membrane as a Barrier to the Free Diffusion of Proteins

A RECENT investigation of the osmotic properties of the nucleus in intact frog oocytes has indicated that the nuclear membrane of these cells is not freely permeable to large molecules (mol. wt. 40,000 or greater)[1]. The present investigation was carried out in order to determine whether or not similar results could be obtained for oocytes of an unrelated species.

Immature oocytes of the *Cecropia* moth (family, Saturniidae), in which yolk formation was just beginning and in which the nucleus was clearly visible, provided a suitable material for this purpose. The ovarioles were removed from animals that had completed one-third of the pupal-adult moult, and placed in a hanging drop of *Cecropia* pupal hæmolymph in a moist chamber. A 0·4 per cent solution of fluorescein-labelled rabbit γ-globulin (fraction II) in nine parts 0·151 *M* potassium chloride (in a sodium chloride solution which is isotonic with this concentration of potassium chloride, *Cecropia* oocytes will neither swell nor shrink: Telfer, W. H., personal communication) and one part 0·01 *M* phosphate buffer was then micro-injected into the cytoplasm of suitable oocytes. The *p*H of the above solution was 7·4; a value selected after injections of phenol red into the oocytes indicated a cytoplasmic *p*H of 7·0 or higher. The protein solution was injected near the nucleus, and care was taken to avoid contact between the micropipette and the nuclear membrane. No changes in the appearance of the cells were observed as a result of micro-injection. 10 min. after being injected, the oocytes were removed from the moist chamber and fixed for 20 min.–2 hr. in three parts 95 per cent alcohol and one part glacial acetic acid. After paraffin embedding, the cells were sectioned at 7 or 8μ. The paraffin was removed from the slides with xylene and the sections were mounted in Schillebar's oil (Grade *A*).

When the slides were observed with the fluorescence microscope, the fluorescence was found to be distributed throughout the cytoplasm. There was, however, no sign of fluorescence in the nucleus (Fig. 1). The results have been interpreted as meaning that the nuclear membrane of *Cecropia* oocyte acts as a barrier to the free diffusion of γ-globulin (mol. wt., approx. 165,000). The main criticism of this interpretation is that, following injection into the cytoplasm, the protein could have been removed from solution either by precipitation or by binding to certain cytoplasmic constituents. However, the fact that the fluorescence was widely distributed through the cytoplasm within 10 min. after the injection indicates that the mobility of the fluorescein-labelled

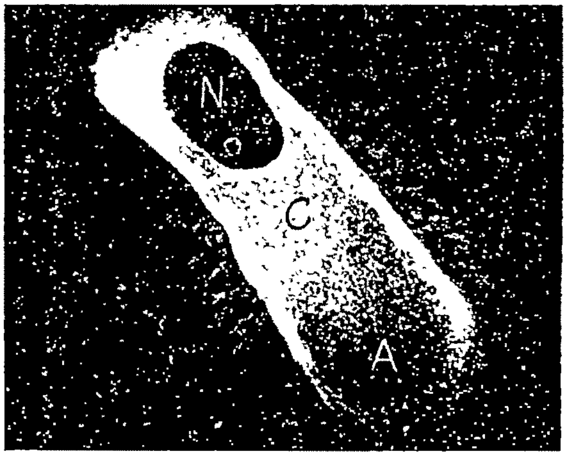

Fig. 1. A section through an injected oocyte showing the absence of fluorescein in the nucleus (*N*). Under the fluorescence microscope the nucleus appeared blue and the entire cytoplasm (*C*) was yellow. Due to a high concentration of granules, the fluorescence in area (*A*) did not show up with black-and-white photography

γ-globulin was sufficient for it to pass through the nuclear membrane if the latter were not impermeable.

It should be emphasized that the rather short 10-min. interval between injection and fixation was chosen for the specific purpose of determining whether the nuclear membrane can act as a barrier to the free diffusion of molecules having a large molecular weight. The inability of a protein to penetrate the nuclear membrane in short-term experiments does not necessarily rule out the possibility that proteins can actually pass into the nucleus. It is not inconceivable, as has been suggested by Stern and Mirsky[2], that an energy-requiring transport mechanism exists whereby molecules which cannot freely penetrate the nuclear membrane can enter the nucleus.

This investigation was supported by funds from a Rockefeller grant to the Zoological Laboratory. We wish to thank Dr. William H. Telfer for supplying the animals and also for his advice and encouragement, and Dr. John Marshall for supplying the fluorescein-labelled γ-globulin and for the use of his fluorescence microscope.

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[1] Harding, C. V., and Feldherr, C. M., *J. Gen. Physiol.*, **42**, 1155 (1959).

[2] Stern, H., and Mirsky, A. E., *J. Gen. Physiol.*, **37**, 177 (1953).

Reaction of Hibernators to Various Low Temperatures of the Environment

IN order to establish the conditions in which hibernators do not maintain their constant body temperature and enter hibernation in summer, the following experiments were made with European hamsters (*Cricetus cricetus* L.) in July and September 1959. Animals were exposed to various low temperatures and their oxygen consumption and rectal temperature recorded for 4 hr. Examples of the results of these experiments are given in Figs. 1 and 2, and mean values of the oxygen consumption in Table 1.

It can be seen that in the experimental temperature − 8° C. the oxygen consumption of the hamsters rose very steeply at the beginning of the experiment to a high level which was maintained throughout the experimental period. Rectal temperature remained constant at the original value about 38° C. The animals behaved like true homoiotherms in these conditions.

In the experimental temperature + 10° C. the course of the changes in metabolism and rectal temperature was quite different. The initial rise of the oxygen consumption was slow and a maximum was not reached until the animals were cooled to some degree. Hereafter the oxygen consumption decreased slowly, but distinctly in a wave-like manner; in connexion with it the rectal temperature fell similarly, the decrease amounting in the mean to 5·2° C. within 4 hr.

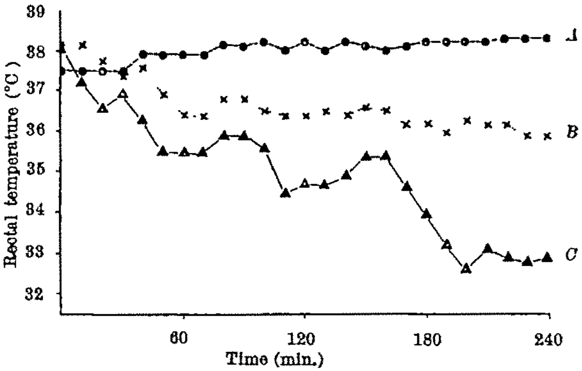

Fig. 1. Typical course of the changes of the rectal temperature in the European hamster in the experimental temperature −8° C. (*A*), + 2° C. (*B*) and + 10° C. (*C*)

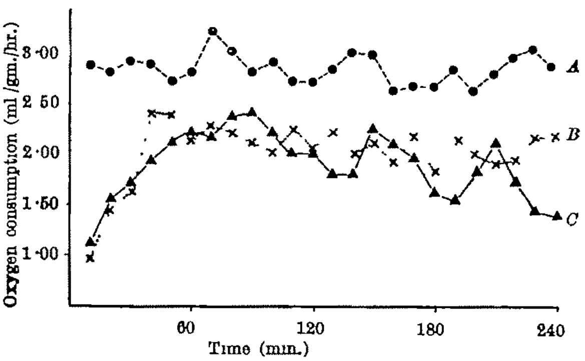

Fig. 2. Typical course of the changes of the oxygen consumption in the European hamster in the experimental temperature − 8° C. (*A*), + 2° C. (*B*) and + 10° C. (*C*)

In the experimental temperature + 2° C. reaction was intermediate between the two cases described: the oxygen consumption rose slowly, though more rapidly than in + 10° C., to a level which was lower than in − 8° C. and remained constant in the further course of the experiment. The rectal temperature fell rather rapidly during the first hour; later its fall was much slower.

The relative instability of the body temperature of hibernators even in periods of activity is well known[1-3]; it may, therefore, be questioned whether the fall of the rectal temperature observed in the experimental temperature + 10° C. after 4 hr. can be taken as evidence of a certain optimum temperature in which hibernation is induced. To obtain further evidence, animals were kept at a low temperature overnight (15 hr.) and their rectal temperature determined at the end of this period. The hamsters tolerated the temperature − 8° C. without any harm, their body temperature remained constant (38·2 ±

Table 1. MEAN VALUES OF OXYGEN CONSUMPTION IN THE EUROPEAN HAMSTER DURING EXPOSURE TO VARIOUS LOW TEMPERATURES OF THE ENVIRONMENT (SIX ANIMALS IN ONE GROUP)

Body-weight (gm.)	Experimental temperature (° C)	Oxygen consumption ml./gm./hr. First hr.	Second hr.	Third hr.	Fourth hr.
295–315	− 8	2·92 ± 0·18	2·90 ± 0·22	2·81 ± 0·29	2·83 ± 0·21
285–315	+ 2	1·80 ± 0·25	2·12 ± 0·14	2·04 ± 0·11	2·03 ± 0·17
290–320	+ 10	1·75 ± 0·18	2·13 ± 0·25	1·83 ± 0·17	1·62 ± 0·09

0·3° C. before, 37·6 ± 0·6° C. at the end of the exposure in six animals) and were normally reactive after 15 hr. In the environmental temperature + 10° C. their rectal temperature fell in the mean to 22·6 ± 3·2° C. (six animals), their mobility was considerably restricted and the hind parts of the body were partly torpid. The animals reacted only weakly to strong sound stimuli; sometimes they gave out throat sounds similar to those produced at the time of arousal from hibernation.

Preliminary experiments with the hedgehog (*Erinaceus europaeus* L.) gave results very similar to those obtained with the hamster.

Our results are analogous to the findings of Deane and Lyman[4] in the golden hamster and show that in hibernators exposed to a high stress a thermoregulatory apparatus of an effectivity comparable to that of non-hibernating mammals can be set in function. The decreased reflex control of warm production[5] appears to manifest itself only within those limits of the environmental temperature which may be taken as a component of the complex of the permissive environmental factors[6].

Although the lethargic state in hibernators has been sometimes experimentally produced in summer[7], cases have not yet been described in which the animals show signs of transition to the lethargic state after such a short exposure to low temperature. The facts described do not as yet exclude the necessity of a specific state of preparation for inducing the hibernation. They favour, however, the view that environmental conditions have the greatest importance.

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[1] Slonim, A. D., "Zhivotnaya teplota i yeyo regulaciya v organisme mlekopitayushikh", Izd. Akad. N. U.S.S.R. (1952).

[2] Johnson, G. E., *Biol. Bull.*, 57, 107 (1929).

[3] Herter, K., "Igel" (Geest and Portig K G., Leipzig, 1952).

[4] Deane, H. W., and Lyman, C. P., *Endocrinol.*, 55, 300 (1954).

[5] Adolph, E. F., *Amer. J. Physiol.*, 166, 92 (1951).

[6] Strumwasser, F., *Amer. J. Physiol.*, 196, 8 (1959).

[7] Eisentraut, M., "Der Winterschlaf mit seinen okologischen und physiologischen Begleiterscheinungen" (VEB Gustav Fischer Verlag, Jena, 1956).

Physiological Mechanisms concerned in the Maintenance of Lactation in the Goat and Sheep

THE anterior-pituitary galactopoietic hormones have been found to be essential for the maintenance of lactation in all species so far studied[1], and it is generally believed that the continued secretion of the galactopoietic complex is entirely dependent on nervous stimuli arising from the mammary gland[2].

Recently, some amplifications of Selye's theory have been put forward, and it has been suggested that the nerve stimuli may effect the release of prolactin and adrenocorticotrophic hormone from the anterior pituitary not directly but by way of the neurohypophysis through the intermediary of oxytocin[3] and perhaps vasopressin[4].

The experiments which gave rise to the neuroendocrine theory of the maintenance of milk secretion were carried out mainly on laboratory animals, and we thought it of interest to investigate whether the same conclusions are generally applicable to other species. For this purpose we have repeated many of these experiments on small ruminants (goats, sheep) and we have started by studying the effects of section of the afferent nerve pathways of the reflex. In a former experiment, the following procedure was carried out in goats and sheep during the first pregnancy.

Section of the lumbar nerves (first, second, third and fourth pairs), section of the perineal nerve with bilateral lumbar sympathectomy. In this experiment the operated animals, after parturition, gave considerable yields of milk.

Since this work was reported[5], we have carried out other experiments involving a greater degree of denervation of the mammary gland:

(1) Severance of all connexion between the mammary gland and the body except the inguinal blood vessels, with perivascular sympathectomy of inguinal blood vessels, followed by suturing of the gland back into position[6].

(2) Section of the spinal cord at *T*11 with bilateral sympathectomy and splanchnicectomy, also removal of the spinal cord between *T*11 and *L*5[7].

(3) Nerve section, resulting in insensitivity of the mammary gland and the adjacent areas, the experimental design and result of which are summarized in Tables 1 and 2. To eliminate any effects of possible regeneration of the perineal nerves, milk yields are given only for the first fifteen weeks of lactation although the yields were recorded over periods of 25–40 weeks. These results entirely confirm our previous experiments[5–7]. In a recent publication, Tverskoi[8] has similarly reported that cord section at *T*11 in the goat does not stop lactation.

Since, as the above findings show, goats and sheep can give substantial yields of milk in the absence of the milk-ejection reflex, it would seem that the milk-ejection reflex is not indispensable for lactation in these species.

The maintenance of the galactopoietic activity of the anterior pituitary in the absence both of the milk ejection reflex and of all mammary sensation may be explained in one of the two following ways. First, given currently accepted concepts of the maintenance of lactation, it may be supposed that some nerve fibres have remained intact, particularly in the deeper portions of the walls of the blood vessels. In this connexion the difficulty of achieving complete denervation is well known. This explanation, however, would only be valid if the fibres persisting in the walls of the inguinal vessels did not enter the main nerves supplying the mammary gland, but reached the spinal cord, at a level above *T*11 or the sympathetic chain above the thoracic ganglia through which the fibres of the splanchnic nerves pass. These assumptions, however, seem unlikely in view of anatomical studies of the nerve supply of the mammary gland[9]. Moreover, one would also have to assume that these hypothetical residual nerve fibres were concerned not with the transmission of stimuli inducing milk ejection or of painful tactile stimuli from the mammary gland, but were highly specialized and concerned entirely with the regulation of the secretory activity of the anterior pituitary. Our results make it difficult to believe that a small number of perivascular nerve fibres would suffice to transmit impulses sufficiently strong to regulate pituitary galactopoietic activity. Secondly, assuming that complete denervation of the udder has been achieved, it would appear to follow that secretion by the anterior pituitary of the galactopoietic complex is under humoral control.

Our experiments thus lead us to believe that the control of pituitary galactopoietic function may be

Table 1. MILK YIELDS OF EWES (FIRST LACTATION)

	Degree of denervation	No. of animals	Mean production per week for the first 15 weeks (ml.)	Total duration of lactation (weeks)
I*	Lumbar nerves 1, 2, 3, 4, lumbar sympathectomy, splanchnicectomy, perineal nerve, all bilateral section	6	4,083 ± 1,230	15·1
II*	Thoracic nerves 12, 13, lumbar nerves 1, 2, 3, 4, lumbar sympathectomy, perineal nerve, all bilateral section	3	4,537	21
III	Thoracic nerves 12, 13, lumbar nerves 1, 2, 3. 4, 5, lumbar sympathectomy, perineal nerve, all bilateral section	6	3,957 ± 997	20
IV	Total of operated animals	15	4,128 ± 1,583	18·7
V	Normal animals	21	4,481 ± 1,820	19·4

* In groups I and II, the lateral femoro-cutaneous nerve may still provide some innervation.

Table 2. MILK YIELDS OF GOATS (FIRST LACTATION)

	Degree of denervation	No. of animals	Mean production per week for the first 15 weeks (ml.)	Total duration of lactation (weeks)
I	Lumbar nerves 1, 2, 3, 4, lumbar sympathectomy, perineal nerve, all bilateral section	11	3,528 ± 1,385	18
II	Thoracic nerves 12, 13, lumbar nerves 1, 2, 3, 4, perineal nerve, all bilateral section	1	5,238	42
III	Thoracic nerves 12, 13, lumbar nerves 1, 2, 3, 4, 5, lumbar sympathectomy, perineal nerve, all bilateral section	3	2,300	46
IV	Thoracic nerves 12, 13, lumbar nerves 1, 2, 3, 4, 5, perineal nerve, all bilateral section	2	3,652	42
V	Lumbar nerves 1, 2, 3, 4, 5, perineal nerve, all bilateral section	2	3,270	23
VI	Total of bilaterally operated animals	19	3,418 ± 1,390	27
VII	Lumbar nerves 1, 2, 3, 4, lumbar sympathectomy, perineal nerve, all unilateral section	7	3,682 ± 1,610	26
VIII	Normal animals	13	6,753 ± 3,300	25

In addition to bilateral denervations, we performed a sympathectomy on inguinal vessels and a section of the spermatic nerve at the lower inguinal ring.

comparable to that envisaged by Fortier[10] in respect of the secretion of adrenocorticotrophic hormone. On this view two main types of stimuli would condition the maintenance of lactation in the normal animal, the relative importance of these two systems varying with the species. First, nervous stimuli would act on the anterior pituitary according to the concepts of Selye[2] or of Benson and Folley[3], the immediate reaction of the organism to the peripheral stimulation being discharge of hormone. Secondly, there may be systemic stimuli associated with the physiological mechanisms of general adaptation of the organism to the demands of the lactating mammary gland for water, metabolites and hormones. These stimuli might act either directly on the anterior pituitary or at the level of the hypothalamus.

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[1] Folley, S. J., in Parkes, A. S., "Marshall's Physiology of Reproduction", 3rd edit., Chapter 20 (Longmans, Green and Co., London, 1952).

[2] Selye, H., *Amer. J. Physiol.*, 107, 535 (1934).

[3] Benson, G. K., and Folley, S. J., *J. Endocrinol.*, 16, 189 (1957).

[4] Benson, G. K., Cowie, A. T., and Tindal, J. S., *Proc. Roy. Soc.*, B, 149, 530 (1958).

[5] Denamur, R., and Martinet, J., *C.R. Soc. Biol., Paris*, 148, 833 (1954).

[6] Denamur, R., and Martinet, J., *C.R. Acad. Sci., Paris*, 248, 743 (1959).

[7] Denamur, R., and Martinet, J., *C.R. Acad. Sci., Paris*, 248, 860 (1959).

[8] Tverskoi, G. B., *Dokl. Akad. Nauk. URSS.*, 123, 1137 (1958).

[9] Linzell, J. L., *Quart. J. Exp. Physiol.*, 44, 160 (1959).

[10] Fortier, C., *Acta Neurovegetativa*, 2, 56 (1952).

Urinary Œstrogens of the Dog

INVESTIGATIONS on naturally occurring œstrogens in the dog are few. So far as urinary œstrogens are concerned, controversial reports are given by authors using biological methods. Helm[1] and Kust[2] report negative findings for both non-pregnant and pregnant animals, while Lesbouyries and Berthelon[3], Finck[4], and Stiasny[5] found evidence for the presence of œstrogens in the urine of pregnant animals. Nilsson[6] reports positive values for non-pregnant bitches. In none of these investigations was the urine hydrolysed before assay.

So far as we know, no attempt has as yet been made towards characterization of the substances by chemical methods. In our laboratory the highly specific and sensitive method of Brown[7] for the chemical determination of œstrogens in human urine has been adopted for research in domestic animals[8]. This communication presents results indicating that by application of this method no significant amounts of œstrone, œstradiols and œstriol could be detected in the urine from male and from pregnant female dogs.

The material consisted of a batch of urine collected from three healthy male dogs, a batch from three healthy females during the last few days of gestation and a single specimen from another pregnant animal.

Two methods of hydrolysis were applied. In the recovery experiment on urines from males, hot acid hydrolysis was applied. A batch of 100 ml. urine was diluted to 200 ml. and boiled under reflux for 1 hr. using 15 vol. per cent hydrochloric acid. 20 μgm. quantities of free œstrogens were added to a parallel sample before acid hydrolysis. Pregnancy urine was submitted to enzyme hydrolysis, using a combined β-glucuronidase-phenolsulphatase preparation (1 ml. of the enzyme preparation contained 100,000 Fishman-units β-glucuronidase and 50,000–80,000 sulphatase-units). To 300 ml. urine was added hydrochloric acid to pH 5·0, and 60 ml. McIlvaine standard buffer (pH 5·0). After the addition of 1 ml. of the enzyme preparation and 100 mgm. of dihydrostreptomycin sulphate, the solution was incubated at 37° C. for 36 hr.

Quantitative determinations were then made on the different samples, using Brown's method[7] with slight modifications[8]. Values less than 1 μgm./l. of urine were considered insignificant.

Table 1. RECOVERY EXPERIMENT ON URINE FROM THREE MALE DOGS

Œstrone		Œstradiol-17-β		Œstriol	
μgm. added	μgm. recovered	μgm. added	μgm. recovered	μgm. added	μgm. recovered
0	0	0	0	0	0
20	8·7	20	7·9	20	11·8

The œstrogens were added *before* acid hydrolysis.

Table 2. ŒSTROGENS IN THE URINE OF PREGNANT BITCHES

Method of hydrolysis	Urine specimen	Œstrone	Œstradiol-17-β	Œstriol
Enzymic	Pooled from three bitches in late pregnancy	0	0	2·2
1 hr. reflux with 15 vol. per cent hydrochloric acid	Fresh from a single pregnant bitch	4·0	0	13·0
	Same urine after storage for 18 months at 4° C.	0	0	0

All values are uncorrected and represent μgm. per litre urine.

Results for male urine in Table 1 indicate that it is practically free from the œstrogens searched for. The recoveries lie well within the limits found by Diczfalusy and Westman[9] for human urine using a similar method.

As shown in Table 2, urine from pregnant bitches appears to contain only trace amounts of the œstrogens searched for. As enzyme hydrolysis, in our opinion, is considerably more efficient than acid hydrolysis, and is not known to cause destruction of liberated œstrogens, these observations can be considered valid. The presence of œstrogens other than those covered by the method applied in the present investigation can, of course, not be excluded. But generally, there seems to be agreement that the œstrogen-level in the urine of the dog is very low, regardless of physiological state. Our results tend to confirm this view.

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[1] Helm, K., *Tierärzt. Rdsch.*, 37, 671 (1931).
[2] Küst, *Deutsch. Tierarztl. Wschr.*, 39, 788 (1931).
[3] Lesbouyries and Berthelon, *Bull. Acad. Vet. Fr.*, 9, 62 (1936).
[4] Finck, *Rec. Med. Vet.*, 112, 628 (1936).
[5] Stiasny, H., *Z. Geburtsh. Gynäk.*, 116, 108 (1937).
[6] Nilsson, S. A., *Skand. Vet. T.*, 38, 643 (1948).
[7] Brown, J. B., *Biochem. J.*, 60, 185 (1955).
[8] Velle, W., "Undersokelser over Naturlig Forekommende Östrogener hos Drovtyggere og Gris", thesis (Oslo, 1958)
[9] Diczfalusy, E., and Westman, A., *Acta Endocrinol.*, 21, 321 (1956).

PLANT PHYSIOLOGY

A Possible Relationship between Nucleic Acid Metabolism and the Initiation of Zygospores of *Rhizopus sexualis*

IT has been observed[1] in the homothallic fungus *Rhizopus sexualis* (Smith) Callen that the initiation and early stages of maturation of zygospores, which at 20–25° C. occur within a few days of inoculation, are completely inhibited at temperatures below 10° C. Later stages in the maturation of zygospores initiated at room temperature and afterwards transferred to low temperature are not inhibited; this suggests that a possible cause of the inhibition is a block in the synthesis of some compound or compounds essential for the early stages of zygospore production.

Some evidence is now available that the first stages of zygospore development are associated with a change in the nucleic acid metabolism of the organism. The nucleic acid content of cultures at different stages of development has been investigated by a procedure based on the perchloric acid extraction method of Ogur and Rosen[2], and it has been found that the level of total nucleic acids (either per culture or per unit dry weight of mycelium) reaches a maximum at the time of zygospore initiation and then falls. By a technique based on that of Korson[3] and used for fungi by Griffiths[4], the ribonucleic acid and the deoxyribonucleic acid of the cells can be differentially stained. This reveals that a rather high concentration of ribonucleic acid is maintained in the hyphal tips, up to the time of zygophore differentiation. Afterwards the level of ribonucleic acid falls rapidly; this fall appears to be accompanied by a rise in the deoxyribonucleic acid content, but later observations are made difficult by the pigmentation of the zygospore wall. In cultures in which zygospore formation is inhibited by low temperature the level of ribonucleic acid remains high. These results suggest that the deoxyribonucleic acid synthesis accompanying zygospore development may be at the expense of the ribonucleic acid of the mycelium, and that the inhibition noted at low temperature is due to a blockage of synthesis of deoxyribonucleic acid. A similar effect of low temperature upon the transformation of ribo- to deoxyribo-nucleic acid in higher plants has long been known[5].

Investigations of the effect of mature zygospores upon further zygospore initiation and development in the same culture resulted in the observation that a volatile compound is produced by mature cultures and is capable of stimulating zygospore production in younger cultures kept at room temperature and, in some instances, of initiating zygospore formation at low temperature. Mature cultures of related species of fungi also produce volatile substances with similar, although smaller, effects on cultures of *R. sexualis*. The activity is not due to carbon dioxide or ammonia, or to a lowering of oxygen tension. The active compound is apparently basic, since it can be removed from a gas stream by scrubbing with dilute hydrochloric acid and is not removed by potassium hydroxide. Attempts to identify the compound by means of a mass spectrometer have been inconclusive, but the fairly frequent occurrence of peaks of mass 15 and 16 is consistent with the possibility that the compound is methylamine. So far as is known, methylamine has not been reported as a metabolic product of *R. sexualis*, but it would not be surprising if it were formed in small amounts, especially in older cultures. When methylamine was supplied at the rate of 1 μgm. per hr. in the air stream passing over cultures at 9° C., there was evidence of a partial reversal of inhibition, in that irregular hyphal swellings similar to pseudophores were induced. In cultures in which zygospore initials were present before chilling, methylamine increased their rate of maturation. Other compounds, such as serine, choline and, to a lesser extent, formic acid, were effective when supplied with the culture medium in increasing zygospore production in cultures in which initiation had already begun. Methionine, betaine and glycine had no effect.

The evidence at present available suggests that the effect of low temperature on zygospore formation is to inhibit the interconversion of ribo- and deoxyribonucleic acid; this inhibition is at least partially reversed by chemicals which are closely associated with the metabolism of methyl and other monocarbon compounds in the cell[6]. In this connexion it is of interest to note that a major difference between ribo- and deoxyribo-nucleic acids lies in the presence of the pyrimidine uracil in ribonucleic acid and its replacement by thymine (5-methyl uracil) in deoxyribonucleic acid. Some of the chief metabolic sources of the methyl moiety of thymine incorporated in the deoxyribonucleic acid of young rats have been found to be serine and choline[7]. It is tempting to speculate that the non-availability of methylating agents needed for the synthesis of thymine and deoxyribonucleic acid may be the chief factor in the low-temperature inhibition of zygospore formation in *R. sexualis*. Critical experiments to test this hypothesis are in preparation, and it is hoped to publish a fuller account of this work when further results are available.

We wish to thank Dr. L. E. Hawker for introducing us to this problem, and for her help, interest and encouragement throughout the work. We are indebted to the Department of Scientific and Industrial Research for a studentship for one of us (P. M. H.).

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[1] Hawker, L. E., Hepden, P. M., and Perkins, S. M., *J. Gen. Microbiol.*, 17, 758 (1957).
[2] Ogur, M., and Rosen, G., *Archiv Biochem.*, 25, 262 (1950).
[3] Korson, R., *Stain Tech.*, 26, 256 (1951).
[4] Griffiths, E. S., *Trans. Brit. Mycol. Soc.*, 42, 132 (1959).
[5] Darlington, C. D., and La Cour, L. F., *J. Genet.*, 46, 180 (1944).
[6] Bach, S. J., *Ergeb. Physiol.*, 48, 529 (1955).
[7] Elwyn, D., and Sprinson, D., *J. Amer. Chem. Soc.*, 72, 3317 (1950).

Effect of Gibberellic Acid on Hypocotyl Growth of Lettuce Seedlings

Gibberellins are physiologically defined by their ability to induce shoot elongation in certain dwarf plants and they have been demonstrated in various plant extracts using dwarf maize plants[1] and dwarf peas[2–4] as assay material. Other assay material includes leaf sections of wheat seedlings[3] and sections of cucumber tendrils[5]. A convenient method employed by Murakami[6] involves the use of rice seedlings. Dwarf maize and pea plants, although very sensitive, are less convenient for assaying the several fractions obtained after paper chromatographic separation of plant extracts. An attempt was made to develop a simple means of bio-assay for this purpose.

The internode elongation shown by dark-grown pea seedlings can be reproduced in light-grown seedlings by gibberellic acid treatment. This has, in fact, been used as a bio-assay technique[7]. In seeds with an epigeal type of germination there is elongation of hypocotyl tissue as well as of internodes in the absence of light and it was decided to investigate the effect of gibberellic acid on hypocotyl growth in three species: *Lactuca sativa* (variety Grand Rapid), *Lepidium sativum*, *Hyoscyamus niger*. There have recently been brief reports[8,9] of the stimulatory effect of gibberellic acid on lettuce seedling growth.

Lettuce seeds were sown in the dark at about 25° C. After 2 days seedlings were selected with radicles of a similar length (6–8 mm.) and planted in 4 in. Petri dishes lined with filter paper moistened with 6 ml. of test solution. 10 seedlings were used per treatment. They were placed 15 cm. below a light source consisting of two daylight fluorescent tubes (5 ft., 80-watt), in which position the temperature was about 28° C. Hypocotyl length was recorded after 5 days, each hypocotyl being measured to the nearest mm. Gibberellic acid had a significant effect on hypocotyl growth at concentrations of 0·1 mgm./l. and above. Growth in 100 mgm./l. gibberellic acid was almost as great as growth of untreated seedlings in the dark. There was a linear relationship between hypocotyl elongation and the logarithm of gibberellic acid concentration. In the dark, hypocotyl growth was affected very little by gibberellic acid treatment. In subsequent experiments measurements were taken after 3 days and seeds were germinated in continuous light. This increased the light inhibition of hypocotyl growth but did not increase sensitivity to gibberellic acid.

Table 1. EFFECT OF GIBBERELLIC ACID ON HYPOCOTYL EXTENSION OF LETTUCE SEEDLINGS

Treatment, gibberellic acid in mgm./l.	Mean hypocotyl length ± *S.E.*	
	A	*B*
Untreated	6·0 ± 0·21	4·2 ± 0·13
0·01	6·0 ± 0·21	4·7 ± 0·21
0·1	9·7 ± 0·45	7·3 ± 0·33
1	13·1 ± 0·43	11·6 ± 0·54
10	17·3 ± 0·36	13·5 ± 0·40
100	19·1 ± 0·55	—
Untreated (dark)	20·3 ± 0·71	—
100 (dark)	21·8 ± 0·74	—

A, Lettuce seedlings germinated in dark prior to treatment. Measurements taken after 5 days. *B*, Lettuce seedlings germinated in light. Measurements taken after 3 days.

To investigate the specificity of the growth-response to gibberellins light-grown lettuce seedlings were also treated with the auxins, β-indoleacetic acid and 2-naphthoxyacetic acid. In neither case was there any positive growth response. Also sucrose had no effect on hypocotyl growth.

Hypocotyls of *Lepidium* and *Hyoscyamus* also showed positive growth responses to gibberellic acid but the effect was less marked than in lettuce.

The following procedure was employed when the lettuce hypocotyl test was used for assaying plant extracts following paper chromatographic separation. The extract was strip-loaded on a 10 cm. wide strip of Whatman No. 1 chromatography paper and developed in a descending fashion with a suitable solvent. The developed chromatogram was cut into ten transverse strips, each strip being cut into two and placed in a Petri dish on filter paper moistened with 6 ml. of water. Two-day-old lettuce seedlings were then planted on the chromatogram strips and hypocotyl growth measured after 3 days.

Using this technique it was possible to detect gibberellin-like activity in plant extracts, although the success of the bio-assay was mitigated by the presence of inhibitory substances in the extract, preventing true quantitative estimates from being made. It is intended to try out further chromatographic techniques which will effect more efficient separation of gibberellins from inhibitory substances.

The chief merit of this lettuce hypocotyl test lies in its simplicity. Seedlings are available for the bio-assay within 48 hr. of sowing and are small enough to be grown directly on the solution under test.

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[1] Phinney, B. O., West, C. A., Ritzel, M., and Neely, P. M., *Proc. U.S. Nat. Acad. Sci.*, 43, 398 (1957).
[2] Radley, M., *Ann. Bot.*, N.S., 22, 297 (1958).
[3] McComb, A. J., and Carr, D. J., *Nature*, 181, 1548 (1958).
[4] Simpson, G. M., *Nature*, 182, 1528 (1958).
[5] Galun, E., *Experientia*, 15, 184 (1959).
[6] Murakami, Y., *Bot. Mag. Tokyo*, 70, 376 (1957).
[7] Phillips, I. D. J., Vlitos, A. J., and Cutler, H., *Contrib. Boyce Thomp. Inst.*, 20, 111 (1959).
[8] Poljakoff-Mayber, A., and Mayer, A. M., *Bull. Res. Counc. Israel*, 7, D, 54 (1959).
[9] Krekule, J., and Ullmann, J., *Biologia Plantarum*, 1, 22 (1959).

IMMUNOLOGY

Inhibition of the Inductive Phase of Antibody Formation by 6-Mercaptopurine examined by the Transfer of Isolated Cells

The inductive phase of antibody formation is a period that differs not only quantitatively but also qualitatively from the later productive phase[1]. This is seen from the effects of X-irradiation[2] and of cortisone[3] on antibody formation, and in particular from the results of tissue-culture experiments[4].

When inhibitors act on the whole organism, however, it is impossible to determine reliably whether they affect the inductive phase only, or also the productive phase. Antigen persists in the tissues for at least a week and during this time the mesenchymal stem cells normally proliferate. This gives rise to a situation in which the newly developing cells are stimulated by antigen (commencement of the inductive phase), while cells which were previously in contact with the antigen are in the productive phase. Since the outcome of the effect of inhibitors on the organism is not instantaneous, it is not always easy to decide during which phase the cells are affected. I therefore examined the effect of inhibitors on a suspension of isolated spleen cells transferred to newly born rabbits[5].

One of the inhibitors selected was 6-mercaptopurine, in an attempt to verify the working hypothesis that ribonucleoprotein, which is responsible for the synthesis of specific protein (antibody) during the productive phase, is itself synthesized during the inductive phase[6]. In earlier experiments 6-mercaptopurine had been used to inhibit antibody formation in adult rabbits immunized with *Salmonella paratyphi B* antigen[7]. Using *S. paratyphi B*, antibody formation was not inhibited even by toxic doses of 6-mercaptopurine, evidently because this antigen evokes a secondary reaction. When Schwartz, Stack and Dameshek[8], however, later used protein antigen ('primary reaction'), they obtained complete inhibition of antibody formation by 6-mercaptopurine.

In the present experiments, 1 ml. spleen cells isolated by the method described in a previous paper[5] was administered intraperitoneally to young rabbits; 1 ml. contained 50 × 10^6 cells. In all the experiments 6-mercaptopurine was administered subcutaneously in amounts of 0·5 mgm./100 gm. body-weight daily.

In the first experiments, cells from normal unimmunized donors, mixed *in vitro* with heat-inactivated *Brucella suis* antigen (7 × 10^8 micro-organism), were administered. In animals which received 6-mercaptopurine on the first five days after cell transfer, antibody formation was completely inhibited. Cells administered without the simultaneous administration of 6-mercaptopurine formed antibodies in the normal manner (Fig. 1).

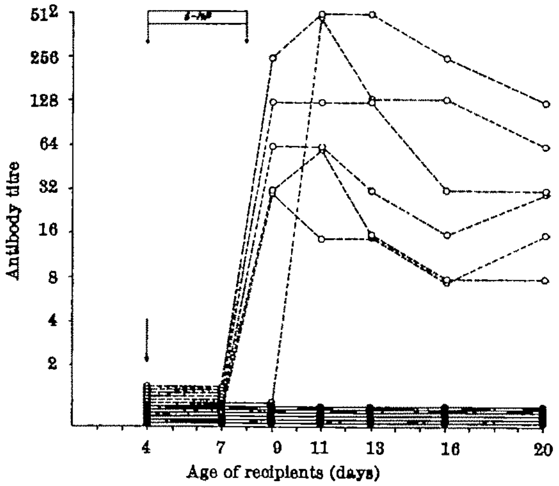

Fig. 1. 6-Mercaptopurine administered simultaneously with transfer of normal cells mixed with *B. suis* antigen *in vitro* (full line). The same cell suspension was administered without 6-mercaptopurine to control recipients (interrupted line)

In other experiments, cells from an immunized donor, that is, already forming antibodies, were transferred. The administration of 6-mercaptopurine in the same doses and at the same times did not prevent these cells from forming antibodies (Fig. 2). These results demonstrate that 6-mercaptopurine does not have an inhibitory effect if antibody formation has already developed (that is, during the productive phase).

In order to determine more precisely the phase of antibody formation as affected by 6-mercaptopurine, this substance was administered at different time-intervals after transfer of normal cells mixed with antigen *in vitro*. When administered 24 hr. after cell transfer, it inhibited antibody formation in every

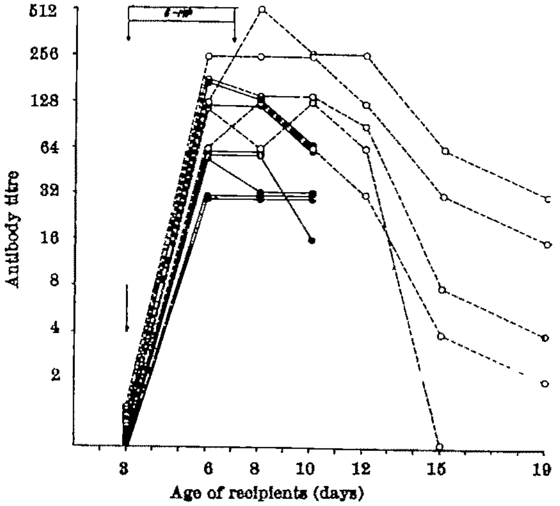

Fig. 2. 6-Mercaptopurine administered simultaneously with cells from previously immunized donor (full line). The same cell suspension was administered, without 6-mercaptopurine, to the controls (interrupted line)

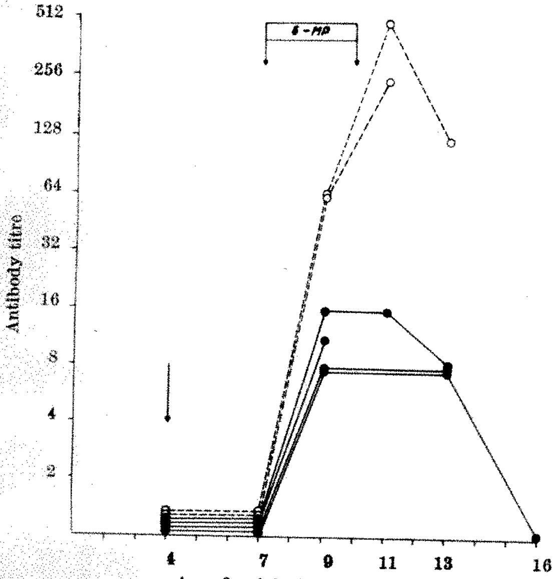

Fig. 3. 6-Mercaptopurine administered 72 hr. after transfer of normal cells incubated with *B. suis* antigen *in vitro* (full line). The same cell suspension was administered, without 6-mercaptopurine, to the controls

case. If it was administered 72 hr. after transfer, the transferred cells formed antibodies, although in low titres only (Fig. 3). Using a bacterial antigen, therefore, the crucial period for sensitivity to 6-mercaptopurine is 48–72 hr. after injection of the cells mixed with antigen *in vitro*; this corresponds exactly to the inductive period, as determined in tissue cultures[4].

It was usually found during these experiments that the dose of 0·5 mgm. 6-mercaptopurine/100 gm., which normally is tolerated well, was lethal for young animals to which cells already forming antibodies were transferred (Fig. 2). This is probably due to summation of the effect of 6-mercaptopurine and the metabolic demands of the transferred antibody-producing cells.

The results show that 6-mercaptopurine is effective during the inductive phase only; the mechanism of its inhibitory action supports the conception that processes during the inductive phase are associated with the synthesis of nucleoprotein. These results form part of a wider series of transfer experiments in which different types of inhibitors (for example, colchicine, ethionine) are used in an attempt to define exactly the conditions and metabolic processes of the inductive phase of antibody formation.

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[1] Šterzl, J., Proc. Symp. Mechanism of Antibody Formation, Prague, 1959 (in the press).

[2] Dixon, F. J., Talmage, D. W., and Maurer, P. H., *J. Immunol.*, **68**, 693 (1952).

[3] Berglund, K., and Fagraeus, A., Atti VI. Congr. int. microb., **2**, 231 (1953).

[4] Šterzl, J., *Experientia*, **15**, 62 (1959); *Folia Microbiol.*, **4**, 91 (1959).

[5] Šterzl, J., *Folia Biol.*, **1**, 193 (1955); **3**, 1 (1957).

[6] Šterzl, J., "Mesenchymal Tissue during Immunisation and Infection" (Praha, 1954).

[7] Šterzl, J., and Holub, M., *Cs. biol.*, **6**, 75 (1957); *Folia Biol.*, **4**, 59 (1958).

[8] Schwartz, R., Stack, J., and Dameshek, W., *Proc. Soc. Exp. Biol. Med.*, **99**, 164 (1958).

BIOLOGY

Myxomatosis in Britain

In his article, Dr. C. H. Andrewes[1] directs attention to the contrast between the host/vector relationship in Britain and Australia. When we have acquired further information about the biology of the rabbit flea (*Spilopsyllus cuniculi*) and also other possible arthropod vectors, we may find that a somewhat different set of factors comes into operation in Britain between major epizootics.

After the initial outbreak of the virulent type of myxomatosis, when the rabbit population at Ashton, Peterborough, was at its lowest ebb, the few survivors which were caught showed a greatly reduced individual flea infestation. It might have been supposed that, owing to the number of dead rabbits from which the fleas had scattered in search of living hosts, the remaining healthy rabbits, especially any immune survivors, would become progressively more heavily infested. But this was not so. All survivors examined six months after the first great epizootic harboured at the most four or five fleas each, quite frequently no fleas at all, or fleas other than rabbit fleas. In the year preceding the outbreak 500 rabbits examined from the same area showed a 100 per cent infestation-rate with a mean per rabbit of approximately 70 fleas on the small sample counted. Three years later, when rabbits were becoming more numerous at Ashton, rabbit fleas were again plentiful, although no actual counts were made at this time.

It therefore seems possible that the chief vectors during a big epidemic may be different from those between, or right at the end, of a big epizootic. Mosquitoes spread myxomatosis, at any rate to a limited extent, in Britain[1]; and it is probable that other winged vectors such as black fly (*Simulium*) are capable of transmitting the disease. During the period of reduced rabbit and flea population the winged vectors may well play the major part as agents of dissemination, and if this were so, the avirulent strains of myxomatosis would, between big epizootics, enjoy certain advantages similar to those described for these strains in Australia. When the rabbit and flea populations build up again, which they do fairly rapidly, the rabbit flea would assume once more its role as chief vector in Britain and consequently there would again be some evolutionary pressure in favour of a virulent virus.

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[1] Andrewes, C. H., Thompson, H. V., and Mansi, W., *Nature*, **184**, 1179 (1959).

Glucose-6-Phosphate Dehydrogenase Deficiency Trait in Nigeria

We were led to look for the presence of this trait in Nigeria because of the demonstration that 10 per cent of American Negroes are deficient in this enzyme[1] and the discovery that hæmolytic anæmia caused by certain drugs (for example, primaquine) develops only in the presence of this sex-linked inherited deficiency of red cell glucose-6-phosphate dehydrogenase[2].

We have applied the rapid screening test as described by Motulsky[3] to 204 male Nigerians of which 139 were Ibo subjects, all of them inmates of Uzuakoli Leprosarium, and 65 were Yoruba subjects attending University College Hospital, Ibadan. Assay for glucose-6-phosphate dehydrogenase[3] was carried out on all 20 persons found deficient by the rapid screening test, and on 18 normal subjects. Our results are shown in Table 1.

Table 1. INCIDENCE OF DEFICIENCY OF GLUCOSE-6-PHOSPHATE DEHYDROGENASE IN RED CELLS OF 200 MALE NIGERIANS

Tribe	No. examined	No. deficient
Ibo	139	9
Yoruba	65	11

The demonstration of the presence of the trait in Nigeria raises several interesting problems: (1) The suggestion made by Motulsky[3] that subjects lacking the enzyme may exhibit a relative resistance to falciparum malaria could be investigated by field-studies of children less than five years of age, and by a study of severely ill malarial children admitted to hospital. (2) The presence of the trait may determine the onset of hæmolytic anæmia induced by dapsone in Nigerian leprosy patients, the incidence of which is approximately 10 per cent (Davey, F., personal communication). (3) The tribal distribution of the trait will have to be assessed in a far larger group of unselected persons. (4) The presence of the trait in adult Nigerians should be looked for in cases of unexplained hæmolytic anæmia often summarily dismissed as 'blackwater fever'.

The co-operation of Drs. F. Davey and L. Hogerzeil in obtaining blood samples is greatly appreciated.

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[1] Beutler, E., *Blood*, **14**, 103 (1959).

[2] Garson, P. E., Flanagan, C. L., Ickes, C. E., and Alving, A. S., *Science*, **124**, 484 (1956). Szeinberg, A., Sheba, C., and Adam, A., *Nature*, **181**, 1256 (1958). Gross, R. T., Hurwitz, R. E., and Marks, P. A., *J. Clin. Invest.*, **37**, 1176 (1958). Childs, B., Zinkham, W., Browns, E. A., Kimbro, R. L., and Torbert, J. V., *Bull. Johns Hopkins Hosp.*, **102**, 21 (1958).

[3] Motulsky, A. G., and Campbell, J. M., *Blood* (in the press).

A New Record of *Mytilicola intestinalis* Steuer, a Parasitic Copepod of Mussels

SINCE the description of *Mytilicola intestinalis* by Steuer in 1902[1] from Trieste, it has been found in various localities of the Mediterranean and appears to have spread from this area to the Atlantic seaboard of Europe and around the British Isles[2]. Infection with this parasite has resulted in serious damage to mussel beds in many areas, with an attendant effect on the mussel industry[3]. Any information regarding the spread of this parasite is consequently of some importance. It is therefore interesting to record the taking of two female *M. intestinalis* in the Indian Ocean–Malacca Strait area, position 7° 12′ N., 97° 12′ E.

The two specimens taken (Fig. 1), approximately 8·5 mm. and 7·0 mm. long, were sufficiently distinctive to refer to this species rather than to either of the two other described species, namely, *M. orientalis* Mori and *M. porrecta* Humes. Dr. Krishnaswamy, of the University of Southampton, has kindly examined the specimens for me and agrees with the identification.

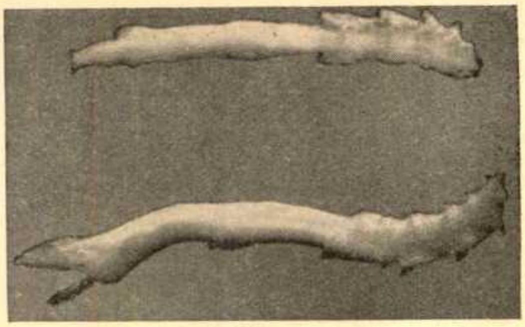

Fig. 1. Adult female *Mytilicola intestinalis*, 8·5 mm. and 7·0 mm. long

The circumstances in which the two specimens were taken were quite remarkable, both being taken in a surface plankton sample, during darkness, while the ship was drifting, hove to. The station was 50 miles outside the 100-fathom line and 67 nautical miles from the nearest land; the depth was 520 fathoms (951 m.).

There is no question of any contamination from European sources, nor is there the possibility that they had been carried from the shore since the water was clear blue and typically oceanic, as was the plankton. The possibility that they had been carried to the surface by currents is considered non-existent. Both my colleague on the cruise and myself are of the opinion that there was no growth of mussels on the ship's hull, although the possibility cannot be ruled out entirely (*M. viridis* L. is the common *Mytilus* of this area). Even if there were infected mussels on the hull it is difficult to explain how the parasites left the host and entered the net, which was some yards from the hull. The possibility that they had been regurgitated by a mussel-eating fish or bird is discounted in view of the excellent condition of the specimens. That adult female *Mytilicola* can be planktonic appears doubtful since the parasite has so far been taken only inside the mussels and never free.

However this may be, the occurrence of this species in the Malacca Strait extends its range of distribution, and increased collection in other areas may well prove the species to be more widely distributed than was hitherto known.

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[1] Steuer, A., *Zool. Anz.*, **25**, 635 (1902).

[2] Bolster, G. C., *Fish. Invest., Lond.*, **18** (6), 30 (1954).

[3] Hepper, B. T., *Fish. Invest., Lond.*, **20** (3), 21 (1955).

First Successful Hybrid between the Two Jute-yielding Species, *Corchorus olitorius* L. (Tossa) × *C. capsularis* L. (White)

ALTHOUGH numerous attempts had been made in the past to cross the two jute-yielding species, namely, *C. olitorius* and *C. capsularis*, no success was achieved[1]. The recent report by Ganesan *et al.*[2] that following hybridization between the two species a few-celled embryo and few free endosperm nuclei are formed encouraged us to repeat the above crosses. The failure to obtain hybrid seedlings has been attributed by these authors to the degeneration of embryo and endosperm at an early stage of their development. An attempt was therefore made to induce the full

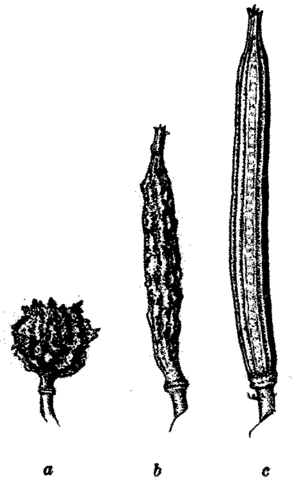

Fig. 1. Fruit shape and size in the hybrid and parents. *a*, Spherical fruit of *C. capsularis*; *b*, hybrid, note the projections and depressions along the ribs; *c*, 10-ribbed elongated fruit of *C. olitorius*, ribs are parallel and smooth. × ⅘

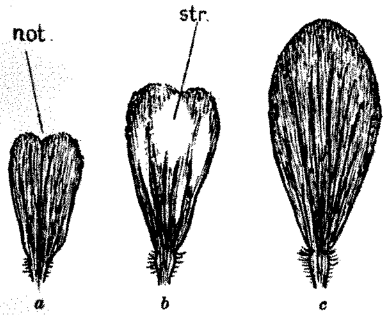

Fig. 2. Petal shape and size in the hybrid and parents. *a*, Notched petal of *C. capsularis*; *b*, notched petal of hybrid with irregular white streaks running from apex to base; *c*, petal of *C. olitorius*. *not.*, notch; *str.*, streak. × 5

development of the embryo and endosperm by the use of hormones. Following pollination, therefore, the pedicels of the flowers were wrapped with cotton which had been previously soaked in 300 p.p.m. 3-indole acetic acid. It was observed that the application of hormone increased the fruit set in the crosses and the seeds obtained therein appeared more developed than in fruits without hormone treatment. Of the 365 seeds obtained from 15 fruits in 115 crosses between *C. olitorius* (♀) and *C. capsularis* (♂), only 7 germinated, and of these only 3 grew to adult plants. No hybrid was obtained in the reciprocal cross.

All the 3 hybrids were slow growing, smaller in height and had a smaller base diameter compared with the parents. The hybrids showed dominance of the female parent in respect of the presence of a pair of stipules, shape of buds and fruits. In the character of leaf shape, B/L ratio of leaf, petal shape, presence of a notch on the upper margin of the majority of petals, irregularly ribbed fruits with alternate projections and depressions running along the ribs, the hybrids showed dominance of the *capsularis* parent (Fig. 1). The hybrids were intermediate in respect of serration of leaf margins, shape of bud and flower, colour and size of petals (Fig. 2), number of stamens and pollen diameter. In addition, the leaves of the hybrids showed areas of yellowish-green colour surrounded by normal green pigment. The petals showed irregular white streaks (Fig. 2*b*) running from the apex to the base surrounded by normal yellow pigment. The presence of these probably indicates the interference in the production of pigmentation brought about by new genic combination. Such disturbance in the production of pigmentation has been reported previously by Harland[3] in the progeny of interspecific crosses of *Gossypium* species, for example, *hirsutum–barbadense* crosses. The first few flowers in the hybrids withered away. Even selfing did not help in the setting of fruits. Almost 50 per cent of the flowers afterwards formed developed into fruits. These fruits varied a great deal in their size and the number of good seeds they contained.

All the 55 back-crosses to the *capsularis* parent failed. One hundred and twenty back-crosses to *olitorius* resulted only in the formation of 7 fruits.

All the diakinetic nuclei studied in the hybrids showed 7 bivalents. One of the 7 bivalents showed incomplete pairing. The two chromosomes in this pair were of unequal length. In all the diakinetic nuclei studied only one pair of chromosomes was found to be attached to the nucleolus. Except for the presence of two univalents in a small number of pollen mother cells, mostly 7 bivalents were seen at metaphase I. Lagging chromosomes at M_{I} and M_{II} were seen in only a small percentage of pollen mother cells. These irregularities were reflected in the presence of nearly 25 per cent sterile pollen grains.

It is known that cryptic structural hybridity obscures the true homology of the parental chromosomes of the hybrid as differences in the small segments of chromosomes do not interfere in the pairing of homologous chromosomes. Whether chromosome pairing in the hybrid was of this type or it really expressed the close homology between the chromosomes of the two species could not be ascertained due to the smallness of chromosomes.

The factor which led to the degeneration of embryo and endosperm at an early stage may have been counteracted by 3-indole acetic acid. The inhibitory effect being removed, the embryo and endosperm developed fully.

The climatic condition and weather in Hyderabad are different from those obtaining in Bengal and this may have also partly contributed to the success of the cross.

The hybrids have opened up new possibilities to jute breeders because by means of selection through a number of generations it may be possible to obtain a pure line with the desirable qualities of both the species combined.

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[1] Patel, J. S., Ghose, R. L. M., and Das Gupta, B., Agric. Res. Mem. No. 3, Indian Central Jute Committee, Calcutta, 1 (1944).

[2] Ganesan, A. T., Shah, S. S., and Swaminathan, M. S., *Curr. Sci.*, **26**, 292 (1957).

[3] Harland, S. C., "The Genetics of Cotton" (London, 1939).

Presence of *Aphelenchoides cocophilus* in the Roots of *Cocos nucifera*, the Coconut Palm

LITERATURE on red ring disease of coconuts, caused by the nematode, *Aphelenchoides cocophilus*[1,2], leaves the reader in doubt regarding the primary site of infection. Nowell[3] referred to the disease in Grenada as 'root disease' and considered the roots to be the primary site of infection. Later[4], he changed his opinion and regarded root infections as secondary to stem. Other workers[5,6] suggested that the disease was transmitted by the palm weevil, *Rhyncophorus palmarum* (L.). One of us[7] advanced the view that the disease frequently started in the base of the tree and suggested that infection might occur via the roots.

While making a series of observations on the disease in the field, we detected the presence of *Aphelenchoides cocophilus* in the roots of three apparently healthy coconut trees. As a result of this, we conducted a survey of a young plantation and examined the roots of 171 trees; root infections were detected in 55 of these trees; two of these root-infected trees were suffering from red ring infection of the stem.

The distribution of the worm was investigated in a number of individual trees and the maximum density was usually found 4–8 ft. away from the bole, and a continuous distribution of nematodes was detected for a distance of up to 14 ft. from the bole. The worms were usually found in the lateral surface roots down to a maximum depth of about 12 in. below the surface; the density of worms in these roots never approached that found in the stem tissues and rarely exceeded 200–300 worms per gm.

Trees, the roots of which were infected, frequently showed symptoms which we considered as indicative of root damage. The most outstanding symptom was a drooping of the bunches of nuts when these were present and a wilting and drooping of the lower leaves: there was no evidence of yellowing. Sometimes when roots of trees showing these symptoms were examined, *Aphelenchoides cocophilus* was not found, but many living specimens of *Rotylenchus* sp. were recovered.

Roots infected with *Aphelenchoides cocophilus* differ from healthy roots in that the root cortex which is firm in texture and white in colour in healthy roots assumes an orange-red coloration and becomes soft and spongy. These symptoms in the roots are not specific to *Aphelenchoides cocophilus* nor, for that matter, to nematode infection, but can also be induced by a variety of other causes.

Preliminary observations suggest that initial infection of roots occurs some distance from the bole, and spreads inwards; but further work is necessary to establish this. The presence of worms in the distal parts of the root system of apparently non-infected trees, when these roots mingle with the infected roots of adjacent trees, suggests the transmission of worms from tree to tree via the root system.

The plantation in question has been observed for four months since root infections were first detected. During this time 7 of the 55 root-infested trees have succumbed to red ring disease; in six of these, the distribution of red ring was consistent with infection starting at the base; the other was definitely the result of an infection starting in the crown. Of the 116 trees in which no root infection was detected only one tree has succumbed; in this, although the characteristic red ring only extended up a distance of 4–5 ft. and there were no petiole infections, yet worms were recovered from the root system for a distance of up to 10 ft. from the bole at one side. The majority of the root-infested trees which succumbed to the disease did so two or three months after root infection was first detected, so it is probable that *Aphelenchoides cocophilus* can remain in the root system for a considerable time before infecting the stem.

It is necessary to emphasize that the observations herein recorded are limited and do not as yet permit the drawing of generalized conclusions.

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[1] Cobb, W. A., *W. Ind. Bull.*, **17** (4), 203 (1919).

[2] Goodey, T., "Plant Parasitic Nematodes" (Methuen, London, 1933).

[3] Nowell, W., *Agric. News*, **17**, 389 (1919).

[4] Nowell, W., *Agric. News*, **18**, 398 (1919).

[5] Martyn, E. B., *Trop. Agric.*, **30** (1, 3), 43 (1953).

[6] Bain, F. M., and Fédon, S. A., "Agronomia Tropicale", July–Sept., 1951 (Maracay, Venezuela, 1951).

[7] Fenwick, D. W., Col. Office Rep. No. 40617, Jan. 1957 (1957).

Effect of Micro-Organisms on the Development of Roots and Root Hairs of Subterranean Clover (*T. subterraneum*)

ALTHOUGH considerable information is available on the rhizosphere microflora and the influence of plants on this population, relatively little is known of the effects of these micro-organisms on the host plant apart from organisms such as *Rhizobium* spp., plant pathogens and mycorrhizas. It has been shown that root growth can be affected by the filtrates of cultures of rhizosphere organisms[1] and some antibiotic substances (polymyxin[2], streptomycin and griseofulvin[3]); however, in all these cases the products of microbial growth rather than the micro-organisms themselves have been added to the roots.

In the course of some general rhizosphere studies being conducted in this laboratory, it was observed that the root systems of plants inoculated with soil suspension appeared stunted and were more easily washed free of adhering sand than corresponding sterile root systems. Some of the results of subsequent studies are reported in this communication.

Seed of subterranean clover was surface sterilized with calcium hypochlorite, germinated on agar and non-contaminated seeds transferred to tubes of autoclaved sand and agar deeps. The sand was in 3 cm. × 20 cm. tubes to a depth of 5 cm. and brought to the equivalent of field capacity with plant nutrient solution[4]; three plants being grown in each tube. The agar deeps were in 1·5 cm. × 25 cm. tubes with 15 cm. of 1 per cent agar prepared with plant nutrient solution; one plant being grown in each agar deep. One half of the tubes were inoculated with 0·1 ml. of a 1-in-10 suspension of pasture soil, the other half receiving 0·1 ml. of a similar soil suspension which had been autoclaved. The tubes were transferred to a controlled-environment room which provided a 12-hr. light period each day of 1,000 ft.-candles on the plants with day and night temperatures of 18° C. and 15° C. respectively. The daily increments in growth of roots in agar were measured, while plants growing in sand tubes were harvested twice each week for root-length measurements and root-hair observations. Twelve replicates of each treatment were set up in the agar series and 12 plants harvested from each of the sterile and non-sterile sand treatments for each observation.

The mean daily root-length increments for sterile and non-sterile clover plants grown in agar (Table 1) show that elongation of the tap root was considerably reduced in the presence of micro-organisms, although there were no indications of root invasion or damage due to pathogens. The standard deviation for the daily measurements shown in Table 1 indicates a

Table 1. MEAN DAILY INCREMENTS IN THE GROWTH OF STERILE AND NON-STERILE SUBTERRANEAN CLOVER ROOTS IN AGAR

Days after planting	Daily increment (mm.) Sterile	Daily increment (mm.) Non-sterile	Days after planting	Daily increment (mm.) Sterile	Daily increment (mm.) Non-sterile
1	4·0 (1·2)*	4·2 (1·6)	12	4·2 (0·66)	1·9 (1·5)
2	3·7 (1·2)	3·0 (0·89)	13	4·4 (0·42)	1·8 (1·1)
3	3·9 (1·4)	2·2 (2·5)	14	5·4 (0·70)	2·0 (1·1)
4	6·0 (1·2)	2·8 (2·2)	15	5·0 (0·72)	1·2 (1·2)
5	7·5 (2·2)	4·9 (2·4)	16	5·5 (0·90)	2·1 (1·1)
6	7·9 (2·5)	5·0 (3·2)	17	4·8 (0·81)	1·9 (1·3)
7	7·1 (1·3)	6·4 (3·7)	18	4·8 (0·63)	1·8 (1·2)
8	6·2 (1·1)	4·7 (3·3)	19	5·0 (0·77)	1·6 (1·4)
9	6·2 (1·6)	4·6 (3·5)	20	5·1 (1·0)	1·6 (1·2)
10	4·0 (0·98)	2·7 (2·2)	21	5·3 (1·1)	1·6 (1·1)
11	5·0 (0·89)	3·6 (3·2)	22	4·8 (0·78)	1·2 (1·1)

* Standard deviation.

much greater variability among the non-sterile plants which may be due to different types and numbers of micro-organisms on the different roots. The stunting effect of micro-organisms was also observed to a lesser degree in plants growing in sand. The growth of the plant tops did not seem to be affected by the micro-organisms on the roots, but this may not be so where the plant nutrients are not in luxury amounts as is the case in Hoagland and Arnon's nutrient solution.

Observations of the root hairs of plants grown in agar and sand showed that these were fewer in number and shorter in length in the non-sterile treatments. Fig. 1 shows the root hairs on the mid-root portions of sterile and non-sterile clover root systems grown in sand for six weeks. The effect was even more marked in the lower root portions—especially near the tip where the non-sterile were completely devoid of root hairs whereas the sterile roots supported large numbers of root hairs.

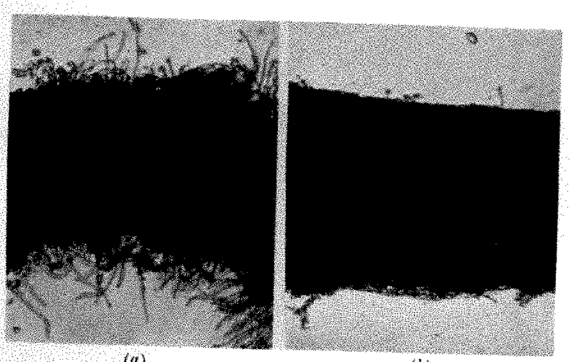
(a) (b)

Fig. 1. Root hair development on the mid-root portions of (a) sterile and (b) non-sterile subterranean clover roots in sand

Similar effects of micro-organisms on root development have also been recorded for tomato, *Phalaris tuberosa* and *Pinus radiata* growing in sand and agar, but the degree of stunting of roots and root hairs varied according to the plant and environmental conditions. This suggests a complex interaction between plants and micro-organisms. A more detailed report of the findings is to be published elsewhere.

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[1] Stolp, H., *Arch. Mikrobiol.*, **17**, 1 (1952).

[2] Norman, A. G., *Arch. Biochem. Biophys.*, **58**, 461 (1955).

[3] Wright, Joyce M., *Ann. Bot.*, N.S., **15**, 493 (1951).

[4] Hoagland, D. R., and Arnon, D. I., Univ. Calif. Agric. Expt. Sta. Circ., 347 (1938).

RADIOBIOLOGY

Conditioned Avoidance Behaviour induced by Low-Dose Neutron Exposure

PREVIOUS studies have demonstrated that exposure to low-intensity γ- or X-rays over a period of 1 hr. or more leads to aversive behaviour in rats[1-4]. The avoidance behaviour is a learned response dependent upon an association between discriminative stimuli and exposure to radiation. The most sensitive test developed so far involves the use of distinctive taste stimuli. Rats will normally consume saccharin-flavoured water in preference to tap water. However, if a rat tastes this solution during a 6-hr. exposure to gamma-rays, resulting in a total dose of 30 r., it will exhibit a marked decrement in preference for saccharin which persists for several weeks post-irradiation. In contrast, an animal which drinks tap water during a similar exposure will maintain the normal preference for the saccharin solution.

The 60-in. cyclotron on the Berkeley California campus of the University of California was employed in this study to determine if fast neutron bombardment would establish avoidance conditioning in rats. The present work differs from previous studies in several important variables of radiation exposure. The animals were exposed for a brief period to fast neutrons at a relatively high dose-rate whereas in previous studies the animals were exposed for several hours to γ- or X-ray radiation at very much lower dose-rates.

Young male rats, approximately 90 days of age, of a Sprague–Dawley strain bred at this laboratory, were used in this study. Litter-mate animals were distributed into control and experimental groups in a balanced design. On three occasions during the two weeks prior to neutron exposure, the animals were processed through sham-irradiation procedures for the purpose of habituating them to manipulative disturbances, which included water deprivation, automotive transportation, confinement to individual irradiation chambers, and consumption of tap water under experimental conditions.

Approximately 24 hr. before neutron exposure, the animals were deprived of water to ensure consumption during the conditioning phase. On the following day they were transported to Berkeley (17 miles) for exposure to the cyclotron beam. Immediately prior to exposure a single plastic bottle was attached to each cage and the animal was allowed to drink during a 20-min. period. As specified by the experimental design summarized in Table 1, the bottles contained either tap water or a saccharin solution of 1 gm. per litre of water. Observations indicated that all the animals tasted the fluid at this time.

For exposure to fast neutrons, each animal was confined to a cardboard cylinder, 2½ in. in diameter by 7 in. in length. The 60-in. cyclotron supplied

Table 1. EXPERIMENTAL TREATMENT FOR EACH GROUP WITH RESPECT TO RADIATION DOSE AND TYPE OF FLUID CONSUMED PRIOR TO EXPOSURE

Group	No. of animals	Neutron dose (rads)	Fluid consumed before exposure
I	14	0	tap water
II	16	0	saccharin solution
III	16	7·5	tap water
IV	15	7·5	saccharin solution
V	14	15	tap water
VI	15	15	saccharin solution

neutrons by bombarding a thick beryllium target with 12 MeV. protons. The mean neutron energy was approximately 2 MeV. The animals were placed at isodose distances, 30 in. from the target. Groups III and IV received 7·5 rads at 10 μamp. beam current while groups V and VI received 15 rads at 20 μamp. Both doses were delivered in 4 min. The animals were turned 180° at the mid-point of exposure to provide a more uniform distribution of dose. Dosimetry for field geometry and exposure dose was made by measuring the fast neutron flux with sulphur threshold detectors as described by Tochilin and Kohler[5]. Gamma background was approximately 12 per cent of the neutron dose under the conditions of exposure used. Groups I and II were sham-irradiated in a corridor outside the water tank shield which surrounds the cyclotron. The environment in the corridor was comparable to that of the radiation exposure room.

The post-irradiation test for conditioned behaviour began approximately 40 hr. after the neutron exposure. The water bottle on the home cage was replaced by two bottles; one containing tap water and the other containing saccharin solution. The amount of fluid consumed from each bottle for a 24-hr. period was measured and the saccharin preference score for each animal is expressed as the ratio:

$$\frac{\text{saccharin solution intake (gm.)}}{\text{total fluid intake (gm.)}} \times 100$$

The results of the saccharin preference test following the neutron exposure are summarized in Table 2. The animals which drank saccharin-flavoured water prior to fast neutron bombardment for 7·5 rads exhibited an 80 per cent reduction in preference for the fluid as compared to the control groups ($P < 0{\cdot}01$). The effect of the 15 rad dose was significantly greater than the 7·5 rad dose ($P < 0{\cdot}01$). Two-thirds of the saccharin group exposed to 15 rads drank less than 1 ml. from the saccharin bottle during the 24-hr. test, but drank normal amounts from their tap water bottle. The animals which consumed tap water before the exposure to fast neutrons drank approximately 90 per cent of their total fluid intake from the saccharin bottle; a preference which was comparable to that of the non-exposed controls.

Table 2. THE MEAN SACCHARIN PREFERENCE SCORE FOR EACH GROUP DURING A 24-HR. TEST COMMENCING 40 HR. AFTER A SINGLE EXPOSURE TO FAST NEUTRONS

Neutron dose (rads)	Groups II, IV, VI (saccharin consumed before exposure)	Groups I, III, V (tap water consumed before exposure)
0	84·6 (4·4)*	90·6 (2·7)
7·5	15·4 (2·7)	89·9 (2·2)
15	4·7 (1·2)	87·3 (3·2)

* Standard error of the mean.

Neutron exposure is apparently an unpleasant experience which the rat associated with the taste of saccharin so that its post-irradiation preference for this substance is altered. The dose employed to establish this avoidance is relatively small; a single total-body dose of approximately 300 rads is required to produce deaths within 30 days post-exposure under the conditions employed in this study.

It may be concluded that fast neutrons as well as γ- or X-rays can be employed as an unconditioned stimulus to produce avoidance behaviour. Furthermore, in contrast to the previous studies which employed radiation exposures of several hours at low dose-rates, the neutron exposure lasted only a few minutes, indicating that the conditioning can be obtained under a wide variety of dose-rates and exposure conditions.

The opinions or assertions contained herein are those of the authors and are not to be construed as reflecting the official views of the Defense Department. We gratefully acknowledge the advice of E. Tochilin and B. W. Shumway concerning neutron dosimetry.

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[1] Garcia, J., Kimeldorf, D. J., and Koelling, R. A., *Science*, **122**, 157 (1955).

[2] Garcia, J., Kimeldorf, D. J., and Hunt, E. L., *Rad. Res.*, **5**, 79 (1956).

[3] Garcia, J., Kimeldorf, D. J., and Hunt, E. L., *Brit. J. Radiol.*, **30**, 318 (1957).

[4] Garcia, J., and Kimeldorf, D. J., *J. Comp. and Physiol. Psychol.*, **50**, 180 (1957).

[5] Tochilin, E., and Kohler, G. D., *Health Phys.*, **1**, 332 (1958).

Estimation of the Cellular Lethal Dose and the Critical Cell Number for the *C3H* Mouse Mammary Carcinoma from Radiosensitivity Studies *in vivo*

IF it is assumed that irradiation of a tumour results in immediate or eventual sterilization of a given proportion of its constituent cells[1], that the magnitude of this effect is an exponential function of dosage[2,3], and that the ability of the residual viable cells to grow progressively depends upon whether their number exceeds that critical value below which their growth is effectively suppressed by host-resistance factors alone[4,5], then the radiosensitivity, or mean lethal dose, of the individual cells, and the critical cell number for the *C3H* adenocarcinoma growing under various conditions, can be estimated indirectly from observed data on the radiosensitivity of the tumour *in vivo*.

Taking the volume of a tumour cell to be of the order of 5×10^{-9} cm.^{3}, and allowing that the stroma and non-viable elements constitute roughly one-half of the tumour mass, we can safely assume each tumour cell to occupy a volume of 10^{-8} cm.^{3}, and accept this convenient unit as a standard 'cell' for the purpose of calculation. The validity of the following argument would not be affected if the actual mean tumour-cell volume differed somewhat from this assumed value.

If the rate of inactivation of tumour cells is exponential with dose, the residual cell population (A) after a given dose of radiation (D) is given by a function of the type:

$$A = N.e^{-kD} \tag{1}$$

where N is the initial cell population, and k a cellular radiosensitivity constant characteristic of the tumour and equal to the reciprocal of the mean lethal dose (synonymous with the 37 per cent-survival dose or D_0).

Table 1 summarizes the results of a series of experiments on the radiosensitivity of the *C3H* mouse mammary carcinoma under various conditions, based upon a total of 799 individual treatments, and representing the most accurate estimates of the relevant parameters currently available[6,7]. The respective median lethal doses for the tumour treated *in situ* are 7,500 rads in immunologically incompetent hosts previously exposed to whole-body irradiation

Table 1. CONSOLIDATED DATA ON THE RADIOSENSITIVITY OF THE *C3H* MOUSE MAMMARY CARCINOMA IN VARIOUS SITUATIONS

Host–tumour relationship	Dose*	Proportion cured	Cure-rate (per cent)	Estimated *LD*50	Standard error	Estimated critical cell No.
Immunologically incompetent hosts	9,000	12/18	67	7,500	± 250	1
	7,500	25/50	50			
Standard isogenic situation	7,500	54/54	> 98	5,660	± 40	25
	6,000	26/39	67			
	5,000	8/42	19			
	4,200	0/108	< 1			
Relatively 'immune' mice	5,000	12/26	54	4,800	± 100	200
	4,200	112/418	27			
	3,500	0/44	< 2			

* Rads: single exposure at 500 rads/min., 250 kVp. unfiltered, 2 cm. field at 25 cm. focus-skin distance, half-value 0·34 mm. copper.

or corticoid hormones[7], 5,660 rads in the standard isogenic host–tumour relationship[6], and 4,800 rads in mice rendered relatively resistant to the tumour by various 'immunizing' procedures[6,8]. In the case of tumour fragments irradiated prior to implantation (revised data[6] not included in Table 1), the dose yielding 50 per cent 'takes' was found to be 3,250 (± 110) rads.

From the data given it appears that the dose required to cure 37 per cent of tumours under conditions of minimal host resistance is about 7,000 rads, at which dose at least 1 viable cell must have persisted, on the average, for each tumour treated, that is, $A \nless 1$. Since these tumours were irradiated when they had reached a diameter of 1 cm. (volume 0·5 cm.³), the initial cell population $N = 0{\cdot}5 \times 10^8$. Substituting in equation (1), it follows that k cannot be greater than 0·00254 rad⁻¹, and that D_0, the cellular mean lethal dose, must be at least 394 rads.

An alternative estimate of this parameter can be obtained from the relative radiosensitivities of tumours (500 mm.³) treated *in situ* in unmodified hosts ($LD50 = 5{,}660 \pm 40$ rads), compared with tumour fragments (1 mm.³) irradiated before implantation ($LD50 = 3{,}250 \pm 110$ rads). If the dosage difference, $\Delta D = 2{,}400$ (± 120) rads, is entirely attributable to the 500-fold difference in cell-population, then $e^{-\Delta D/D_0} = 1/500$, and $D_0 = 385$ (± 20) rads. Since the two estimates are in close agreement, one could take the mean lethal dose for *C3H* carcinoma cells to be of the order of 390 rads.

From the foregoing data, the critical cell number, just permitting continued tumour growth in the mouse, can be computed. The median tumour lethal dose of 5,660 rads for 500 mm.³ growths treated *in situ* corresponds to a residual viable cell population of $0{\cdot}5 \times 10^8/e^{(5{,}660 \pm 40)/390} = 25$ (± 3) 'cells'. Similarly, for the 1 mm.³ fragments irradiated prior to implantation, the dose of 3,250 rads corresponds to a viable cell population of $10^5/e^{(3{,}250 \pm 110)/390} =$ 25 (± 7) 'cells'. A reasonable estimate, therefore, of the critical cell number, permitting 50 per cent growths in the normal *C3H* mouse bearing the standard isogenic mammary tumour, would be of the order of 20–30 cells.

In the group of relatively 'immune' mice, the estimated *LD*50 of 4,800 rads corresponds to a surviving population of more than 200 'cells'.

The cell-population concept, however, cannot in itself account fully for the variation in cure-rate with dosage. Given the average cell population for tumours treated at a particular dosage, factor A in equation (1), the proportion of cured tumours (P) presumed to contain less than Z cells (the critical number), is given by a Poisson exponential summation:

$$P = e^{-A} \sum_{x=0}^{z-1} \left(\frac{A^x}{x\,!}\right) \qquad (2)$$

Comparison of this function with the slope of the empirical 'probit' regression for the data shows the latter to be significantly greater. This difference reflects the variance of the observations, attributable in the main to the inherent dosimetric error, accounting for a 10 per cent coefficient of variation, and in part to minor genetic variation among individual mice.

In summary, the minimum number of cells capable of progressive growth in the three types of host–tumour relationship studied, that is, in the immunologically incompetent, the normal, and the 'immune' states, are of the order of 1, 25 and 200 tumour cells respectively. Assuming a constant radiosensitivity of the cells *per se*, the dose of radiation which is just sufficient to deplete the tumour cell population to the critical level will vary with different degrees of host resistance. Changes in radiosensitivity associated with various host–tumour combinations can, therefore, be accounted for in terms of differences in the corresponding critical cell assemblies.

The possibility that a low oxygen tension had raised the apparent lethal dose in our experiments cannot be excluded, although there is evidence that anoxia was not complete and that a similar degree of oxygenation existed both in the tumour treated *in situ* and in the fragments irradiated before implantation (unpublished data). Since complete anoxia may increase the inactivation dose by a factor of about 3[1], it seems unlikely that our estimate of D_0 would need to be reduced by much more than a factor of 2. On the other hand, consideration of the relative biological efficiency of the relatively soft radiation used in our experiments (half-value layer, 0·34 mm. copper) might necessitate an upward revision of the estimated D_0. Thus the mean lethal dose for the *C3H* carcinoma appears to be substantially greater than that reported for certain other tumours by more direct methods[2,3].

The foregoing reasoning is naturally based upon a quantal response to irradiation, namely, the lethal effect on tumour cells. It fails to account for the 'immunizing' effects induced by radiation-attenuated isogenic inocula, or for the graded reactions and interactions appearing in many tissues, organs and animals, which have been collectively described in terms of 'antigenic equivocation'[8]. It seems probable that the 'equivocation' and 'critical assembly' hypotheses are complementary. A detailed report on these experiments will be published elsewhere.

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[1] Scott, O. C. A., "Advances in Biological and Medical Physics", **6**, 121 (Academic Press, New York, 1958).

[2] Puck, T. T., and Marcus, P. I., *J. Exp. Med.*, **103**, 653 (1956).

[3] Hewitt, H. B., and Wilson, C. W., *Nature*, **183**, 1060 (1959); *Brit. J. Cancer*, **13**, 69 (1959).

[4] Revesz, L., *J. Nat. Cancer Inst.*, **20**, 1157 (1958).

[5] Klein, G., *Cancer Res.*, **19**, 343 (1959).

[6] Cohen, A., and Cohen, L., *Brit. J. Cancer*, **7**, 231, 452 (1953); **8**, 303, 313, 522 (1954); **9**, 600 (1955); **10**, 312 (1956); **11**, 563 (1957); **12**, 561 (1958).

[7] Cohen, L., and Cohen, A., *Brit. J. Radiol.*, **32**, 18 (1959).

[8] Cohen, L., and Cohen, A., *Nature*, **183**, 692 (1959).

An Autoradiographic Study of the Distribution in Mice of Œstrogens labelled with Carbon-14 and Tritium

WE have examined the distribution in mice of the metabolic products of two œstrogens by an autoradiographic method which allows histological localization. It is of interest to use a 'physiological' dose, which Dodds[1] estimates to be close to 1 μgm. hexœstrol in the rat. Glascock and Hoekstra[2], using tritium-labelled hexœstrol in the goat and the sheep, found that the maximum quantity of the injected product which could be recovered did not at any time exceed 0·2 per cent in the uterus or 0·05 per cent in the vagina.

In preliminary experiments, we used œstrone-16-^{14}C of specific activity 2·7 μc./mgm., and in a second series œstradiol-17B-6,7-^{3}H (from C. E. Frost Co., Montreal, and the New England Nuclear Corp., Boston, respectively, and supported by a grant from the Fondation Fontaine), of specific activity 3·85 mc./mgm. The doses were respectively 1 mgm. œstrone-16-^{14}C (2·7 μc.) dissolved in poppy-seed oil and 50 μgm. œstradiol-17B-6,7-^{3}H (200 μc.) dissolved in olive oil, per animal. Female mice, weighing 25–30 gm., were injected at arbitrary stages in the œstrous cycle. The animals were killed after ½, 1½, 4 and 20 hr. in the first series, and after 3, 4, 6 and 12 hr. in the second. Fragments of the uterine tubes, the kidneys and the liver, and the whole of the ovaries were taken out. After fixation for 4 hr. in 10 per cent formaldehyde, two series of biopsies were prepared: the first was washed in water for 1 hr., the other for 12 hr. Because paraffin embedding was unsuitable, owing to the solubility of œstrone and œstradiol in alcohol, sections (20μ) were cut from material frozen with carbon dioxide. The sections were spread on gelatin-covered slides and treated for track autoradiography with nuclear emulsion Ilford *G* 5[3]. They were exposed for 6–10 days, developed and then stained with 'Unna'. Uninjected mice and mice injected with the solvent oils were treated similarly.

The results of the first series were entirely negative (œstrone-16-^{14}C). We observed no localization of β-tracks which could be attributed to the injected material. This failure is not surprising considering the small amount of radioactive material injected (2·7 μc.) and the fact that œstrogens are rapidly metabolized and excreted by the kidneys and the bile ducts[4].

In the second series (œstradiol-17B-6,7-^{3}H) when the animal had been killed after a short time (3–4 hr.) a localization of grains was observed in the kidneys and in the uterine tubes. These grains have the characteristic appearance of those produced by tritium β-rays. In the uterus, these grains are found in the cells of the endometrium and in the lumen of the glandular tubes, in contact with the apical pole of cells (Fig. 1), from which they are not removed by prolonged washing. However, Rengler[5] reports that, in rats injected with œstrone-16-^{14}C, the major portion of the radioactivity of the fluid from washings of the uterine lumen is present as a water-soluble component and that no radioactive steroids can be recovered from this fluid. Such a localization of grains was not observed after 6–12 hr.

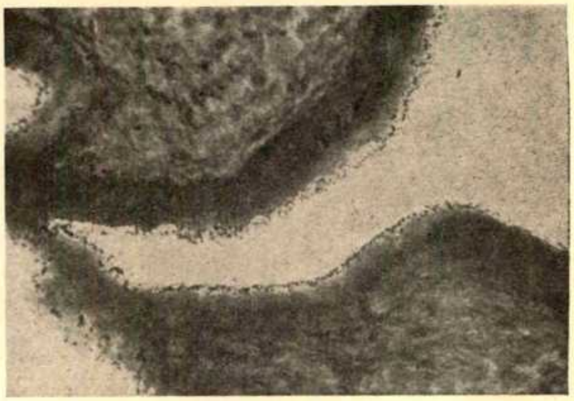

Fig. 1. Grains in the endometrial cells and in the lumen of the glandular tubes

In the kidneys, the grains are found in the periglomerular space and in the first part of the convoluted tubule, after 3–4 hr. (Fig. 2); but they are eliminated by prolonged washing. Similarly, after 6–12 hr., they are scarcely observable. This radioactive material, localized in the first part of the nephron, is therefore probably soluble, and seems to be filtered through the glomeruli. Budy[4] reports that the concentration of radioactive product recovered from the kidney is higher than one would expect, judging from the radioactivity of the urine; she attributes this result either to an effect of the blood, or to a re-absorption by the convoluted tubule. Our results favour the latter view.

In the liver and ovaries we have not observed any distribution of radioactivity which could be correlated with the structure of the tissues. It has been known for a long time that the liver plays an important part in the metabolism and the excretion of the œstrogens, and the absence of systematic localization might be related to the special histological structure of this organ, or to the moment when the animal was killed.

In the ovary, the lack of radioactivity in the present experiments is more difficult to understand, since Glascock[2] has found in this organ, for the same weight, half the radioactivity observed in the uterus.

The control sections were negative. One must, however, point out the presence of artefacts—fine grains and lines where the sections are disrupted.

In conclusion, the histological localization of the metabolic products of tritium-labelled œstrogens seems to be possible. Investigations with a more 'physiological' dose and examination within 2 hr. of injection should not be without interest.

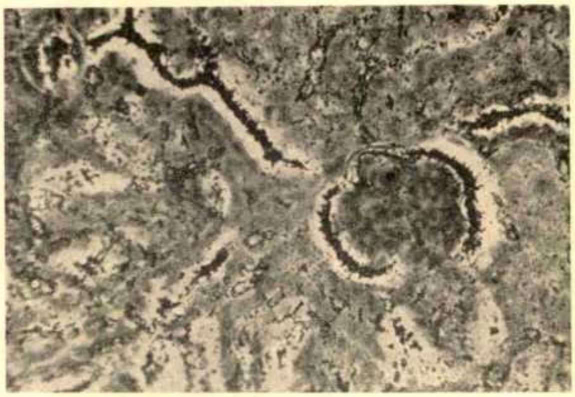

Fig. 2. Numerous grains in the periglomerular space and in the first part of the convoluted tubule

I wish to thank Prof. J. Brachet, who allowed me to study this question in his laboratory, and I am particularly indebted to Dr. A. Ficq for her advice and kind technical co-operation.

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[1] Dodds, E. C., Folley, S. J., Glascock, R. F., and Lawson, W., *Biochem. J.*, 68, 161 (1958).

[2] Glascock, R. F., and Hoekstra, W. G., *Biochem. J.*, 72, 673 (1959).

[3] Ficq, A., *Arch. Biol.*, 66, 509 (1955).

[4] Budy, A. M., *Arch. intern. Pharmacodynamie*, 103, 485 (1955).

[5] Rengler, L., *Proc. Soc. Exp. Biol. Med* , 95, 506 (1957).

BACTERIOLOGY

Interspecific Hybridization among *Mycobacteria*

Experiments conducted in the first part of this work showed that a high degree of streptomycin resistance can be transmitted to *M. phlei* when a streptomycin-sensitive strain of this micro-organism and a streptomycin-resistant *BCG* strain of *M. tuberculosis* are cultured together in 'Tween' liquid medium.

Resistance to 50 μgm. streptomycin per ml. (S^r) was selected as marker in the *BCG* strain; the markers selected for *M. phlei* were pigment formation (Pig^+) and the capacity for growth on plain agar medium (Ag^+). The *BCG* strain may thus be described as S^r Pig^- Ag^-, while *M. phlei* is S^s Pig^+ Ag^+. Colonies of the new organism were picked from a selective medium: tryptose agar containing 50–1,000 μgm. streptomycin per ml. on which neither parent organism could grow when planted alone. Their inability to grow on this medium in 30 or more days served as control. The frequency of resulting streptomycin-resistant organisms was estimated to be 1–10 per 10^6 *M. phlei* units. Similar results were obtained in four experiments.

One of the strains obtained in the course of these experiments deserved special attention as it resembled the so-called 'atypical' acid-fast chromogenic *Mycobacteria*. This hybrid, in addition to high streptomycin resistance, showed yellow pigmentation (both in the dark and in the light) which, in contrast to that of the parental *M. phlei*, turned orange on prolonged cultivation. At 37° C. the strain grew on tryptose agar, as well as on 'Tween' agar or Loewenstein's medium within 5–12 days; at room temperature growth appeared slowly, in 12–20 days. No cord formation was seen. A high degree of pleomorphism and instability of traits other than the selected markers were found to be characteristic.

Since streptomycin was the only selected marker of the *BCG* strain used in these experiments, it was not possible to make any recombination analysis, or to assess the importance of a cell-to-cell contact (conjugation) in the transfer of streptomycin resistance.

Experiments were, therefore, carried out in order to shed more light on the mechanism of transfer: *M. phlei* was grown in the culture filtrate of the *BCG* strain; and *BCG* in the culture filtrate of *M. phlei*. Cell-free filtrates of *M. phlei* were never observed in these experiments to bring about changes in the *BCG* strain. Sterile culture filtrates of the *BCG* strain, on the other hand, conferred the streptomycin resistance to *M. phlei* cells. This could be repeated in three experimental series. The frequency, however, was 10^2 lower, that is, 1–10 per 10^8 *M. phlei* units. These findings indicate that the transfer of streptomycin resistance does not necessarily require cells as donors, and thus transduction may be the actual mechanism of transfer. (In the trials already carried out deoxyribonuclease treatment of the filtrate has not affected significantly its activity.)

These experiments, both those in which a *BCG* culture and those in which cell-free filtrates of a *BCG* culture were used as donors, disclosed also that: (1) The streptomycin resistance of descendant organisms increased to a level of more than 1,000 μgm. per ml. as against a level of 50 μgm. per ml. of the parental *BCG* strain of *M. tuberculosis*. (2) This high level of streptomycin resistance was attained in a single step, while the parent strain of *BCG* acquired the much lower resistance in four or five distinct steps. (3) In addition to this, a considerable portion of the resulting variants showed on isolation dependence on streptomycin. (4) Evidence was also obtained supporting the assumption that the transfer of hereditary traits is uni-directional, that is, from *BCG* strain of *M. tuberculosis* to *M. phlei*.

The results of the work described here appear to bear a resemblance to the findings made in the course of investigations of genetic transfer in various species of bacteria[1-5]. Studies on genetic transfer together with certain recently reported observations with respect to mutation[6-9] may, it is hoped, some day give an answer to the origin of atypical mycobacteria.

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[1] Zinder, N. D., and Lederberg, J., *J. Bacteriol.*, 64, 679 (1952).

[2] Luria, S. E., and Burrows, J. S., *J. Bacteriol.*, 74, 461 (1957).

[3] Leidy, G., Hahn, E., and Alexander, H. E., *J. Exp. Med.*, 104, 305 (1956).

[4] Miyake, T., and Demerec, M., *Nature*, 183, 1586 (1959).

[5] Terada, M , Abstr. Seventh International Congress of Microbiology, Stockholm (1958).

[6] Tarshis, M. S., *Amer. Rev. Tubercul.*, 78, 921 (1958).

[7] Tirunarayanan, M. O., Vischer, W. A., and Bruhla, H., *Schweiz. Z. Path. Bakt.*, 21, 964 (1958).

[8] Mayer, R. L. Eisman, P. C., Gisi, T. A., and Konopka, E. A., *Amer. Rev. Tubercul.*, 77, 694 (1958).

[9] Di Marco, A., Ghione, M., and Zavaglio, V., *Giornale di Microbiologia*, 1, 266 (1956).

Role of *Hfr* Mutants in $F+ \times F-$ Crosses in *Escherichia coli* K12

Genetic recombination between mutant strains of *Escherichia coli* strain *K*12 is governed by a system of mating types[1,2]. The first two mating types to be discovered were called *F*+ and *F*−. In crosses the *F*+ parent donates genetic material to the *F*− parent. The progeny are predominantly *F*+. At the same time it was discovered that the *F* factor was infectious, that is, if *F*+ and *F*− strains were mixed and the organisms of the *F*− strain re-isolated, a proportion had become *F*+. It was therefore possible that the *F*+ state of the progeny was secondary and that any *F*-zygotes became infected by other *F*+ cells.

Then came the discovery of mutant strains of a new mating type known as high-frequency recombining (*Hfr*) strains[1,2]. Whereas *F*+ strains crossed with *F*− strains gave only one recombinant per

Table 1. DETAILS OF FOUR EXPERIMENTS ON SECONDARY INFECTION BY *F* FACTOR AFTER RECOMBINATION

Experiment	*F*− cells per plate	*F*+ cells per plate	Recombinant colonies per plate in first half	Percentage of colonies in first half *F*+	*Hfr* cells added per plate for second half	Recombinant colonies per plate in second half	Percentage of colonies in second half *F*+	Calculated percentage of zygotes in first half originally *F*+
1	2.5×10^8	2.5×10^7	n r	95	2.5×10^3	n r	36	92
2	2.5×10^8	2.5×10^6	25	80	10^4	600	14	77
3	2.5×10^8	2.5×10^7	150	77	10^3	2,000	44	60
4	2.5×10^8	2.5×10^6	4	96	10^4	250	21	95

n r., not recorded

10^5 parents, these *Hfr* strains when crossed with *F*− strains gave about 1,000 times this number of recombinants. The progeny of such crosses were *F*−. *F*+ organisms apparently mutate to the *Hfr* state and *Hfr* clones can be isolated from *F*+ cultures[4].

Jacob and Wollman suggested that these mutant forms were the agents in recombination in *F*+ × *F*− crosses, and that the *F*− progeny so formed were then secondarily infected by the *F*+ cells present. In support of their theory they measured the fluctuation in the number of recombinants obtained from several small *F*+ cultures. This suggested that the recombination was at least in part due to clones of *Hfr* cells. However, this type of evidence does not prove conclusively that cells of such clones are the only donors of genetic material in an *F*+ culture, and Lederberg has suggested that they are not[5]. Some experiments were designed to show whether secondary infection can, in fact, account for the high proportion of *F*+ progeny found in an *F*+ × *F*− cross.

In these experiments we have used *K*12 mutants 58–161/*F*+ (requiring methionine), *W*677/*F*− (requiring threonine, leucine and aneurin) and *Hfr* strains isolated from 58–161/*F*+. Washed log-phase broth cultures of these strains were suspended at approximately $2-5 \times 10^8$ viable organisms/ml. Each experiment consisted of two halves. In the first half the 58–161/*F*+ suspension was mixed with the *W*677/*F*− suspension in a proportion varying from 1 : 10 to 1 : 100. Aliquots of 0·5 ml. of this mixed suspension were immediately spread over each of several plates of minimal agar plus aneurin. The liquid was very quickly soaked up. For the second half of the experiment small numbers of *Hfr* cells were added to the mixed suspension before dispensing on to the plates. The actual numbers are given in Table 1. Both series of plates were incubated 36 hr. and the number of colonies counted. Neither parent could grow on this medium and the colony counts gave an indication of the recombination-rate.

Single colonies from these plates were streaked out on the same medium to obtain colonies free of parental cells and single colonies from these streaks tested for their mating type by crosses with *W*677/*F*− (streptomycin resistant). Those which would cross were recorded as *F*+ and those which did not as *F*−. In experiments 3 and 4 the colonies scored as *F*− were shown not to be chance contaminants by their ability to cross with 58.161/*F*+ (streptomycin resistant) but not with *W*677/*F*− (streptomycin resistant).

In each experiment the addition of the *Hfr* suspension increased the recombination-rate several-fold and therefore most of the progeny in the second half, having come from *Hfr* parents, must have been originally *F*−. The proportion of *F*+ progeny in the second half gives us, then, the rate of secondary infection under these experimental conditions. If it is assumed that the probability of secondary infection is the same in both halves of the experiment we can calculate the proportions of the zygotes in the first half which were originally *F*− and *F*+. This is given in the last column.

While prior mutation to *Hfr* undoubtedly accounts for some of the recombinants in an *F*+ cross, the proportion which are formed via *Hfr* mutation is not certain.

The existence of a significant proportion of *F*+ zygotes in the first half of the above experiment makes it evident that the genetic recombination in an *F*+ × *F*− cross is not mediated entirely by stable *Hfr* mutants of the type so far isolated, but that some *F*+ cells cross directly with *F*− cells.

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[1] Lederberg, J., Cavalli, L. L., and Lederberg, E. M., *Genetics*, **37**, 720 (1952).

[2] Hayes, W., *J. Gen. Microbiol.*, **8**, 72 (1953).

[3] Hayes, W., Cold Spring Harb. Symp Quant. Biol., **18**, 75 (1953).

[4] Jacob, F., and Wollman, E. L., *C.R. Acad. Sci., Paris*, **242**, 303 (1956).

[5] Lederberg, J., Abstracts, Seventh International Congress of Microbiology, Stockholm, p. 58 (1958).

Synthetic Polypeptide Derivatives with Anti-Bacterial Activities

A NUMBER of polypeptides composed of various basic amino-acids have been prepared by Katchalski *et al.*, and it had also been stated that agglutination of Gram-positive and Gram-negative bacteria was caused by these polypeptides, and bacteriostatic or bactericidal activity was observed depending on their concentration[1]. Stahmann and his co-workers[2] have reported that the multiplication of some viruses was restrained by poly-L-lysine.

From these observations and the fact that a number of naturally occurring antibiotics (for example, tyrocidine and polymyxins) have peptide structures and basic characters, it may be assumed that the microbiological activity of these compounds depends fundamentally on these structures and characters.

The synthetic polypeptides composed of basic amino-acids, however, cannot be employed for pharmacological use for they are soon disintegrated by the proteolytic enzymes of the higher organisms.

As previously stated[3], polymethyl-α-poly-L-glutamate can be converted with ethylenediamine into a water-soluble peptide derivative ('Polyanthin') which is resistant to proteolytic enzymes of allantoic liquid and at the same time efficiently hinders the development of bacteriophages T_5, T_6 and T_7. Its subcutaneous toxicity is about $LD50 = 850$ mgm./kgm. in mice.

The possible anti-tuberculous activity of this product has been investigated as follows (the 'Polyanthin' used in the experiments was kindly supplied by the Chinoin Chemical and Pharmaceutical Works Co., Budapest):

(1) To 1 ml. of a solution of 3,000γ 'Polyanthin' 0 01 mgm. (wet weight) of a three-weeks-old *H*37*Rv* culture was added. One part of the suspension was stored at room temperature, the other part at 37° C. Samples of 0·1 ml. were taken at different intervals and inoculated on Löewenstein–Jensen and Sula media. Other samples of 0·1 ml. were injected into pairs of guinea pigs. The culture media were examined after six weeks incubation at 37° C., while the animals were submitted to section and examined after eight weeks. . Both results are summarized in Table 1.

Table 1. Relation of 'Polyanthin' Activity to Time and Temperature of Application against *Mycobacterium tuberculosis*

No. of experiment	Time (hr.)	Temperature (° C.)	No of colonies	Degree of tuberculosis at autopsy
I*a*	16	37	many	generalized
I*b*	16	room	many	generalized
II*a*	24	37	many	generalized
II*b*	24	room	many	generalized
III*a*	72	37	10	moderately generalized
III*b*	72	room	many	moderately generalized
IV*a*	120	37	5	Only changes in regular lymphatic glands, and a few tubercules in spleen
IV*b*	120	room	4	
Control*	120	37	many	generalized
Control	120	room	many	generalized

* The controls were suspensions of tubercle bacilli in physiological saline solution.

The results show that 'Polyanthin' exhibits a bacteriostatic activity, increasing with the time of application.

(2) Thirty guinea pigs (about 350 gm.) were infected subcutaneously with 0·001 mgm. (wet weight) suspensions of *H*37*Rv*-Sula culture. (The suspension was prepared with rust-proof steel balls.) To each of 20 guinea pigs, once, in a single dose, a neutral solution of 60 mgm. 'Polyanthin' was given subcutaneously on the day following the infection. Ten guinea pigs were left for control. After 93 days the animals were submitted to autopsy. The degree of tuberculous change found in the different organs was estimated in whole numbers in the range of 0–4, and mean values were calculated. The following organs were examined: regulare lymphatic gland, retroperitoneal lymphatic gland, spleen, liver and lungs. The index for the control group was 3·4 and for the other group that got the 'Polyanthin' injection 1·4.

These experiments seem to show that 'Polyanthin' exhibits considerable anti-tuberculous activity. Further experiments are in progress.

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[1] Katchalski, E., Bichowsky-Slomnitzki, L., and Volcani, B. E. *Biochem. J.*, **55**, 671 (1953).

[2] Burger, W. C., and Stahmann, M. A., *Arch. Biochem. Biophys*, **39**, 27 (1952).

[3] Kovács, K., Dénes, G., Kótai, A., and Polgár, L., *Naturwiss*, **42**, 628 (1955).

Cystine as an Essential Amino-Acid for the Growth of a Strain of *Shigella dysenteriae* I (*Sh. shiga*)

In the studies on the nitrogen requirements for the growth of members of the genus *Shigella* in a chemically defined medium with various nitrogen sources added singly[1] a strain of *Shigella dysenteriae* 1 was found to grow only when cystine was present.

The basal medium consisted of 5 ml. each of salt solutions *A* and *B*[2], glucose 0·1 *M* and nicotinamide 10^{-5} *M*. To this was added, singly, the following: ammonium chloride (1 per cent), DL-glutamic acid, DL-phenyl alanine, DL-histidine, DL-lysine, DL-threonine, glycine, l-cystine, l-cysteine hydrochloride, DL-leucine, l-arginine, DL-aspartic acid, DL-alanine, DL-tryptophan, DL-valine, DL-*iso*leucine, DL-methionine, DL-tyrosine, DL-proline and DL-serine. The various nitrogen sources were dissolved in the basal medium with gentle heating. Cystine required the aid of hydrochloric acid and for tyrosine *N* sodium hydroxide was needed[3]. The amino-acids were present at 0·01 *M* concentration, except cystine and tyrosine, which were used at 0·0005 *M* and 0·002 *M* concentrations, respectively. The final *p*H of the medium was adjusted to 7·2–7·4; the medium was sterilized by autoclaving at 115° C. for 15 min. The inoculum of the organism was 0·1 ml. of an overnight peptone water growth diluted one in a hundred in sterile double-distilled water. Incubation was carried out aerobically at 37° C. and continued for seven days. If growth occurred during this period, one loopful of the growth was transferred to a second tube of the same medium. The nitrogen source was recorded as giving positive growth if three serial sub-cultures showed growth of the organism.

Cystine was the only amino-acid, among the nitrogen sources tested, in which growth of the strain could be obtained. As cysteine did not produce growth, further experiments were conducted and the results are shown in Table 1. The possibility of impurities in the cystine originally used was explored by using a preparation from another source of supply. The result was as before. The possibility that autoclaved cysteine, by producing colloidal sulphur and colloidal copper sulphide[4], may have caused inhibition of growth was eliminated by using filtered cysteine[5]. An attempt to grow the organism in cysteine containing medium which had been shaken aerobically at 37° C. for 6 hr., in order to abolish inhibition of growth by exogenous cysteine[6], was unsuccessful. There is a possibility that insufficient amounts of cystine were formed from oxidation of cysteine, for in a medium containing 0·02 mgm. of cystine per ml., as used by Proom in the study of the nutrition of Providence strains[7], the strain of *Sh. dysenteriae* 1 did not show any growth.

Johnson[8] suggested that cystine had a nutritional role for a strain of *Sh. dysenteriae* 1 investigated by him. The results of the present investigation in

Table 1. Growth of *Sh. dysenteriae* 1 in Medium containing Cystine and Cysteine

	Medium containing inorganic salts, glucose, nicotinamide and the following	Growth
1	Cystine, 0·0005 *M*	+
2	Cysteine, 0 01 *M*	−
3	Cysteine, 0·0005 *M*	−
4	Cysteine, 0·01 *M* – Medium Seitz filtered	−
5	Cysteine, 0 0005 *M* – Medium Seitz filtered	−
6	Glutamic acid, 0 01 *M* + cystine, 0·0005 *M*	+
7	Glutamic acid, 0 01 *M* + cystine, 0 02 mgm. per ml.	−
8	Glutamic acid, 0 01 *M* + cysteine, 0 01 *M*	−
9	Glutamic acid, 0 01 *M* + cysteine, 0 0005 *M*	−

Table 2. GROWTH OF *Sh. dysenteriae* 1 IN MIXTURES OF AMINO-ACIDS WITH AND WITHOUT CYSTINE. CYSTINE, WHEN PRESENT, WAS AT 0 0005 *M*, ALL OTHER AMINO-ACID CONCENTRATIONS WERE 0 01 *M*

	Medium consisted of basal medium and the following amino-acid mixtures .	Growth
1	Glut. + meth. + tryp. + asp. + cys.	+
2	Glut. + meth + tryp. + asp.	–
3	Glut. + meth + tryp + cys.	+
4	Glut. + meth. + tryp.	–
5	Glut. + asp. + tryp. + cys.	+
6	Glut. + asp. + tryp.	–
7	Glut. + asp. + meth. + cys.	+
8	Glut. + asp. + meth.	–
9	Asp. + meth. + tryp. + cys.	+
10	Asp. + meth. + tryp.	–
11	Glut. + tryp. + cys.	+
12	Glut. + meth. + cys.	+
13	Glut. + asp. + cys.	+
14	Meth. + tryp. + cys.	+
15	Glut. + tryp.	–
16	Glut. + meth.	–
17	Glut. + asp.	–
18	Glut. + cys.	+
19	Meth. + tryp.	–
20	Tryp. + asp.	–
21	Tryp. + cys.	+

Glut., glutamic acid , meth., methionine ; tryp , tryptophan , asp , aspartic acid , cys , cystine.

showing that growth of the strain occurred only when cystine was present in mixtures of amino-acids, as tabulated in Table 2, would also favour an essential nutritional role for the amino-acid for this particular strain of *Sh. dysenteriae* 1.

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[1] Sen, R., Ph.D. thesis, University of London (1959).

[2] Snell, E. E., and Strong, F. M., *Indust. Eng. Chem.*, Anal. Ed., 11, 346 (1939).

[3] Fildes, P., Gladstone, G. P., and Knight, B. C. J. G., *Brit. J. Exp. Path.*, 14, 189 (1933)

[4] Woiwod, A. J., *J. Gen. Microbiol.*, 10, 509 (1954).

[5] Rowatt, E , *J. Gen. Microbiol.*, 17, 297 (1957).

[6] Roberts, R. B., Abelson, P. H , Cowie, D. B., Bolton, E. T., and Britten, R. J., "Studies of Biosynthesis in *Escherichia coli*", Carnegie Institution of Washington Pub. 607 (Washington, D.C , 1957).

[7] Proom. H.. *J. Gen. Microbiol.*, 13, 170 (1955).

[8] Johnson, R. M., thesis, University of New Mexico, University Micro-Films, Ann Arbor, Michigan, Pub. No. 13586 (1955).

VIROLOGY

Persistence of Herpes Simplex Virus in HeLa Cells

PERSISTENCE of herpes virus in cells cultured *in vitro* might provide a suitable model for investigating the phenomenon of latent herpes infection in humans. During a study on the interaction between herpes virus and HeLa cells, it was observed that an extensive cytopathic effect eventually resulted from inoculation of a monolayer culture with the *HFEM* strain of herpes virus[1,2]. A few isolated cells sometimes survived, however, and if growth medium containing antiviral serum was added, colonies grew from these surviving cells, leading eventually to a large population. These recovered cells were apparently healthy and grew as well as the original HeLa cells : there were no gross morphological differences such as those described by Vogt and Dulbecco[3], who studied cells recovered after poliomyelitis infection.

Despite continued growth in the presence of high-titre antiserum, however, the recovered cells were found to carry herpes virus for at least nine months, involving 29 subcultures. The presence of the virus was indicated by the appearance of typical microscopic foci of infected cells or plaques, from which virus could be isolated. These plaques appeared whether or not there was antiserum in the medium. When antiserum was present, however, the plaques remained localized and did not increase in number. It was therefore possible to deduce that there was one plaque-forming unit (presumably an infected cell) in approximately 10^5 cells. If antiserum was removed from the medium, the foci became larger and eventually confluent, with release of infective virus into the medium. No infective virus has been isolated from medium or cells at times when no plaques were visible.

By taking a small population of recovered cells it was possible to grow a sub-line which did not give rise to plaques spontaneously, even in medium free from antiserum. This sub-line was compared with the original HeLa cells for susceptibility to virus recovered from a single plaque in the recovered cells and then grown in chick cells to form a virus stock. With a large dose of virus, the virus-free recovered cells underwent a confluent cytopathic effect with release of virus. Comparative infectivity titrations in the recovered cells and the HeLa cell stock indicated, however, that 10–100 times as much virus had to be present to infect a recovered cell as that required to infect a stock HeLa cell.

The mechanism of persistence of herpes virus in recovered cells has yet to be elucidated ; but the susceptibility of these cells to virus, although reduced, does not suggest a situation resembling temperature bacteriophage, and it is also unlikely that auto-interference of the type studied by Henle *et al.*[4] could be the explanation. It is possible that persistence is due simply to the very slow spread of virus in the selected population of less-susceptible cells in the presence of inhibitory medium. This would resemble persistence of poliomyelitis virus in partially resistant HeLa cells, studied by Ackermann and Kurtz[5] and Vogt and Dulbecco[3]. Unlike the poliomyelitis-infected cultures, however, the recovered cells continued to carry herpes virus in the presence of strong antiviral serum. This is in keeping with the known ability of this virus and the related *B* virus of monkeys to spread between contiguous cells in antiserum[6,7].

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I am indebted to Prof. M. G. P. Stoker for stimulating suggestions and for his interest in this work

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[1] Ross, R. W., and Orlans, E., *J. Path. Bact.*, 76, 393 (1958).

[2] Stoker, M. G. P., Soc. Gen. Microbiol. Symposium on Virus Growth and Variation, 142 (1959).

[3] Vogt, M., and Dulbecco, R., *Virol.*, 5, 425 (1958).

[4] Henle, G., Deinhardt, F., Bergs, V. V , and Henle, W., *J. Exp. Med* , 108, 537 (1958).

[5] Ackermann, W. W., and Kurtz, H., *J. Exp. Med.*, 102, 555 (1955).

[6] Black, F. L., and Melnick, J. K., *J. Immunol.*, 74, 236 (1955).

[7] Stoker, M. G. P., *Brit. Med. J.*, 1, 963 (1957) ; *Nature*, 182, 1525 (1958).

FORTHCOMING EVENTS

(*Meetings marked with an asterisk * are open to the public*)

Monday, January 25

Institution of Electrical Engineers, London Graduate and Student Section (at Savoy Place, London, W.C.2), at 6.30 p.m.—Mr. M. Paskins: "Transistors in Switching Circuits".

Radar and Electronics Association (at the Royal Society of Arts, John Adam Street, Adelphi, London, W.C.2), at 7.30 p.m.—Mr. J. C. G. Gilbert and Mr R. S. Roberts: "The Problems of Technical Reviewing".*

Tuesday, January 26

University College (in the Anatomy Theatre, Gower Street, London, W.C.1), at 1.15 p.m.—Dr. R. W. R. Rutland "Radioactivity in the Earth's Crust".*

University of London (at Queen Mary College, Mile End Road, London, E.1), at 1.30 p.m.—Prof. J. D. Bernal, F.R.S.: "The Liquid State".*

University of London (at the Imperial College of Science and Technology, South Kensington, London, S.W.7), at 1.30 p.m.—Mr. Ernest Gellner: "The Barbarian Scientist ?"* (Further lectures on February 2, 9, 16 and 23.)

Royal Society of Arts, Commonwealth Section (at John Adam Street, Adelphi, London, W.C.2), at 5.15 p.m.—Dr. R. Maclagan Gorrie. "Irrigation and Population in Pakistan, India and Ceylon".

Institute of Marine Engineers (at the Memorial Building, 76 Mark Lane, London, E.C.3), at 5.30 p.m.—Mr. R. Anscomb and Mr. F. Hutber: "Control and Instrumentation of Marine Reactors".

University of London (at King's College, Strand, London, W.C.2), at 5.30 p.m.—Prof. J. Brachet (Université Libre de Bruxelles). "The Rôle of Ribonucleic Acid in Protein Synthesis".*

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. R. J. V. Pulvertaft: "Auto-Immune Reaction".* (Fifth of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on January 28, February 2, 4, 9, 11, 16, 18, 23, 25 and March 1.)

Society of Chemical Industry, Plastics and Polymer Group (at 14 Belgrave Square, London, S.W.1), at 6.30 p.m.—Dr. J. F. Buckley: "Polymerization and Copolymerization of Itaconic Acid and its Derivatives".

Society of Instrument Technology (at Manson House, 26 Portland Place, London, W 1), at 7 p.m.—Symposium on "Flame Failure Detection". Mr. D. H. Surgeoner: "A Review of Current Flame Failure Devices"; Mr. D. Richards: "Applications of Flame Failure Equipment".

Wednesday, January 27

Geological Society of London (at Burlington House, Piccadilly, London, W.1), at 2 p.m. and 5 p.m.—Symposium on "Geological Implications of Recent Geophysical Work in the British Isles".

British Computer Society, London Branch (at the Northampton College of Advanced Technology, St. John Street, London, E.C 1), at 2.30 p.m.—Dr. G. G. Macfarlane. "Storage Elements for Very High Speed Computers".

Institution of Electrical Engineers, Electronics and Communications Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. A. Clow: "The Oral Presentation of Scientific Material".

University of London (at King's College, Strand, London, W.C.2), at 5.30 p.m.—Prof. J. Brachet (Université Libre de Bruxelles) "The Rôle of the Cell Nucleus in Ribonucleic Acid and Protein Synthesis".*

Challenger Society (at the Linnean Society, Burlington House Piccadilly, London, W.1), at 5.45 p.m—205th Scientific Meeting. Mr. W. H. Dawbin: "Whales and Whaling in the South Pacific Ocean".

Institution of Mechanical Engineers (at 1 Birdcage Walk Westminster, London, S.W.1), at 6 p.m.—Dr. J. J. McMullen "Technical and Economic Aspects Covering the Ocean Transportation of Liquid Methane" (Thomas Lowe Gray Lecture).

British Institution of Radio Engineers (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 6.30 p.m.—Dr. K. R. Sturley and Mr. A. E. Robertson: "Training for Operating and Maintaining Television Broadcasting Equipment".

Thursday, January 28

Challenger Society (joint meeting with Representatives from the Marine Laboratories (Development Commissioners' Scheme), in the Lecture Theatre, British Museum (Natural History), Cromwell Road, London, S.W.7), at 10.30 a.m. and 2.30 p.m.—Scientific Papers.

Institution of Mining Engineers (at the Institution of Naval Architects, 10 Upper Belgrave Street, London, S.W.1), at 11.15 a.m.—66th Annual General Meeting.

Linnean Society of London (at Burlington House, Piccadilly, London, W.1), at 5 p.m—Mr. Wm. T. Stearn: "How Many Species of Flowering Plants have Taxonomists Yet to Describe ?"; Dr. W. S. Lacey: "A Botanical Visit to Central and South Africa".

Mineralogical Society (at the Geological Society of London, Burlington House, Piccadilly, London, W 1), at 5 p.m.—Scientific Papers.

Institution of Electrical Engineers (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. A. W. Lines "Radio Communications by Means of Satellites".

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Dr. R. G. White: "Immune Cellular Reactions".* (Sixth of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on February 2, 4, 9, 11, 16, 18, 23, 25 and March 1.)

University of London (at the School of Pharmacy, 29–39 Brunswick Square, London, W.C.1), at 5.30 p.m.—Prof. P. Blanquet (University of Bordeaux). "Ion Exchange Resins and Thyroid Metabolism". (Further lecture on January 29.)

Friday, January 29

Institution of Electrical Engineers, Electronics and Communications Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Mr. R. B. Dyott and Mr. C. R. Russell: "Beam-Type Parametric Amplifiers—Some Aspects of Design and Use".

University College (in the Eugenics Theatre, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. E. L. Lehmann (University of California) "The Efficiency of Non-Parametric Tests". (First of two Special University Lectures on Statistics. Further lecture on February 1.)*

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m—Mr. B. S. Shenstone: "Engineering Aspects of Man-Powered Flight".

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Prof. Edgar Wind: "The Academy of Leonardo Da Vinci".

Saturday, January 30

London County Council (at the Horniman Museum, London Road, Forest Hill, London, S.E.23), at 3.30 p.m.—Dr. G. H. Francis: "Exploration in the Southern Andes of Peru".*

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Research Officer or Research Cadet (with an honours degree in agricultural science, and preferably special training or experience in soils research) in the Soils Division (Soil Physics Department)—The Director, An Foras Talúntais, 33 Merrion Road, Dublin (January 29).

Lecturer and an Assistant Lecturer (honours graduate in physics or chemistry) in Textile Chemistry—The Registrar, The Manchester College of Science and Technology, Manchester 1 (January 30).

Senior Technician in the new Department of Medicine, to take technical charge and assist with biochemical research—The Secretary, Royal Free Hospital, Gray's Inn Road, London, W.C 1 (February 1)

Research Fellow in the Department of Physics, for theoretical work in connexion with investigations now proceeding in the Department into the properties of solids at extreme infra-red and submillimetre wave-lengths—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1 (February 6).

University Demonstrator (with an appropriate honours degree and preferably postgraduate research or teaching experience) in Animal Physiology—The Secretary, School of Agriculture, The University, Downing Street, Cambridge (February 6).

Junior Lecturer in Physiology—The Registrar, Trinity College, Dublin (February 8).

Lecturer or Assistant Lecturer in Statistics—The Registrar, The University, Leeds 2 (February 8).

Lecturer (preferably with a veterinary qualification) in Pharmacology in the School of Veterinary Medicine—The Registrar, Trinity College, Dublin (February 8).

Lecturer of Assistant Lecturer in the Department of Genetics, to assist chiefly with biometrical and population genetics—The Deputy Registrar, The University, Edgbaston, Birmingham 15 (February 10)

Assistant Lecturer or Lecturer (with special interests in inorganic chemistry) in Chemistry—The Registrar, The University, Sheffield (February 13).

Senior Lecturer or Lecturer in the Department of Electrical Engineering (Electronics)—The Registrar, The University, Liverpool, quoting Ref. CV/T (February 13).

Chair of Physics—The Registrar, The University, Southampton (February 15).

Lecturer or Assistant Lecturer in Pure Mathematics—The Secretary and Registrar, The University, Southampton (February 15).

Lecturer or Assistant Lecturer (with a first-class degree in mathematics and a strong interest in research in some field of applied mathematics) in Applied Mathematics—The Secretary and Registrar, The University, Southampton (February 15).

Scientific Officer/Senior Scientific Officer (Physical Chemist) (with a first- or second-class honours degree in physical chemistry or equivalent), at Warren Spring Laboratory, D.S.I.R., Stevenage, Herts, to work on application of radio isotopes in research problems being pursued in the laboratory—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting reference F.872/9A (February 15).

Chair of Botany at Westfield College—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 19).

Chair of Chemistry at Westfield College—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 22).

Chair of Mathematics at Westfield College—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 23).

Scientific Officer (with a first- or second-class honours degree in chemistry) in the Department of Pedology, Forest Soils Section, for duties mainly concerned with studies on nitrogen in peat in relation to afforestation, as well as the development and application of methods for foliage analysis—The Secretary, The Macaulay Institute for Soil Research, Craiglebuckler, Aberdeen (February 27).

Assistant Lecturer or Lecturer in Organic Chemistry—The Secretary and Registrar, The University, Southampton (February 29).

Imperial Chemical Industries Research Fellows in Biochemistry, Chemistry, Engineering, Pharmacology or Physics—The Secretary of University Court, The University, Glasgow (February 29).

Lecturer or Assistant Lecturer in Organic Chemistry—The Secretary and Registrar, The University, Southampton (February 29).

Assistant Lecturer in Chemistry—The Secretary, The University, Exeter (March 1).

Assistant Lecturer (preferably with an interest in theoretical plasma physics) in Physics—The Secretary, The Queen's University, Belfast (March 1).

Lecturer (with experience in one of the following fields . general physical metallurgy, metallurgy of surfaces, high temperature materials and ceramics) in the Department of Physical Metallurgy—The Deputy Registrar, The University, Edgbaston, Birmingham 15 (March 1).

Imperial Chemical Industries Research Fellows in Chemistry, Civil Engineering or Physics—The Registrar, Trinity College, Dublin (March 7).

Imperial Chemical Industries Fellows for research in Physics, Chemistry, Colloid Science, Biochemistry, Engineering, Metallurgy, Pharmacology, Chemotherapy, or related subjects—The Secretary-General of the Faculties, The University, The Old Schools, Cambridge (April 30).

Imperial Chemical Industries Research Fellows in Biochemistry, Chemistry, Chemotherapy, Engineering, Metallurgy, Pharmacology or Physics—The Academic Registrar, University of London, Senate House, London, W.C.1 (April 30).

Turner and Newall Research Fellow in Engineering, Inorganic Chemistry or Physics—The Academic Registrar, University of London, Senate House, London, W.C.1 (April 30).

Agricultural Officer (Irrigation) (with a university degree in agriculture and preferably with experience in tropical irrigation research) in Kenya, to take charge of an irrigation research station and to be responsible for a research programme on irrigated crops and pastures—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.63/7/04/T.

Associate Professor or Assistant Professor (theoretical physicist, preferably with experience and main interest in the field of nuclear theory), to teach courses at the senior and postgraduate levels and to carry on research—The Head, Department of Mathematical Physics, University of Manitoba, Winnipeg, Manitoba, Canada.

Farm Manager (aged 25 to 35 years, with a university degree in agriculture) at Yundum Experimental Farm, Gambia, for technical and administrative duties in running an agricultural experimental station—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD/63/12/05/T.

Librarian—The Secretary, Oxford University Institute of Statistics, St. Cross Road, Oxford.

Master to teach Physics in the Upper School, including the Sixth Form—The Headmaster, St. Dunstan's College, London, S.E.6.

Organic Chemist (with a first- or good second-class honours degree and a good general knowledge of organic chemistry, and preferably postgraduate experience in steroid chemistry), to undertake the synthesis of labelled steroids—The Personnel Officer, The Radiochemical Centre, U.K.A.E.A , Amersham, Bucks, quoting Ref. No. 11/1959.

Research Assistants (2) (with a degree in chemistry or equivalent) in the Department of Chemical Technology, one for work on synthesis of nucleotides and analogous compounds and their biological application in cancer chemotherapy, and the other for work involving the synthesis of isotopically labelled organic compounds and the use of modern physical methods of analysis—The Registrar, Bradford Institute of Technology, Bradford 7.

Research Worker (with experience of research in the field of radioactivity), to be in charge of a newly established Isotope Unit—The Secretary, Queen Elizabeth College (University of London), Campden Hill Road, London, W.8

Senior Lecturer (with an honours degree or equivalent in mechanical engineering together with industrial experience) in the Department of Aircraft Propulsion—The Recorder, The College of Aeronautics, Cranfield, Bletchley, Bucks.

University Extension Lecturers (qualified archæologists specializing in the earlier prehistoric periods) in Archæology—The Deputy Director (Extension), University of London, Senate House, London, W.C.1.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Other Countries

Agricultural Research Council A Bibliography of Farm Buildings Research, 1945–1958 Part 1 Buildings for Pigs. Pp 36. (London: Agricultural Research Council, 1959.) 2*s*. 6*d* [3011

Commonwealth Agricultural Bureaux. Plants Recorded as Resistant to Root-Knot Nematodes (*Meloidogyne* spp.). Catalogued by Dr. Mary T. Franklin and David J. Hooper. Pp. iii+33. (Farnham Royal . Commonwealth Agricultural Bureaux, 1959.) 7*s*. 6*d*. [3011

Forestry Commission Forest Record No 40 · Preliminary Yield Table for Poplar. By J M. Christie. Pp. 16. 1*s*. 6*d*. net. Forest Record No. 42 Use of Home-Grown Softwood in House Construction. By J. R. Aaron. Pp. 12. 1*s*. 3*d*. net. (London: H.M. Stationery Office, 1959.) [3011

Henry Wiggin and Co., Ltd. Publication No. 1122: Welding Nimonic Heat-Resisting Alloys for Gas-Turbine Assemblies. (Based on an article in *Aircraft Production*, July, 1959.) Pp. 19. (Birmingham: Henry Wiggin and Co., Ltd., 1959.) [3011

Government of Northern Ireland: Ministry of Agriculture. Leaflet No. 82: The Production of Clean Milk. Pp. 26. (Belfast: Ministry of Agriculture, 1959.) [3011

Department of Scientific and Industrial Research. Coalfield Papers of the Geological Survey of Great Britain No. 2 The Economic Geology of the Stirling and Clackmannan Coalfield, Scotland; Area South of the River Forth. By W. A. Read. With Palaeontology by R. B. Wilson. Pp. vi+78+2 tables and 15 figures. (Edinburgh and London: H.M. Stationery Office, 1959.) 7*s*. net. [3011

University of Leeds. Imperialism and Colonialism. By Prof. Raymond Aron. (Seventeenth Montague Burton Lecture on International Relations.) Pp. 18. (Leeds: The University, 1959.) 6*d*. [112

Henry Wiggin and Co , Ltd. Publication No. 1529A: Wiggin Nickel Alloys v. Caustic Alkalis. Pp. 51 (11 plates). (Birmingham: Henry Wiggin and Co., Ltd , 1959.) [812

Ten Years of Development in Cyprus: 1949–1959. Pp. 32. (London: Cyprus Government London Office, 1959.) [312

Other Countries

International Atomic Energy Agency. Nuclear Science Fellowships: Information—Opportunities—Procedures. Pp. 24. (Vienna: International Atomic Energy Agency, 1959.) [3011

Commonwealth of Australia: Department of Supply. Australian Defence Scientific Service: Defence Standards Laboratories. Report No. 242: The Laboratory Development and Evaluation of a Thixotropic Protective Oil for Critical Metal Surfaces. By E. C. Kuster. Pp. i+17. (Maribyrnong, Victoria: Defence Standards Laboratories, 1959.) [3011

Report on the Geology of the Leeward and British Virgin Islands. By P. H. A. Martin-Kaye. Pp iii+117+14 figures. (Castries, St. Lucia: Government Geologist, 1959.) [3011

Annual Report of the Calcutta School of Tropical Medicine and the Carmichael Hospital for Tropical Diseases, 1957–58. Pp. 146 (Calcutta: Calcutta School of Tropical Medicine and Carmichael Hospital for Tropical Diseases, 1959) [3011

Fiskeridirektoratets Skrifter. Serie Havundersøkelser. Vol. 12, No. 4: A Note on the Effect of Electric Fields on *Gadus virens*. By Olav Dragesund and Helge Leivestad. Pp. 9. Vol 12, No. 5: Scale Character and Growth Rate: a Study on Small Fat Herring from North Norway. By M. Soeprapto. Pp. 16. (Bergen: A. S. John Griegs Boktrykkeri, 1959.) [3011

Smithsonian Miscellaneous Collections. Vol. 139, No. 6: A Revision of the Silurian Bryozoan Genus *Trematopora*. By Richard S. Boardman. Pp. 14+2 plates. (Publication 4383.) (Washington, D.C.: Smithsonian Institution, 1959.) [3011

Dominion Museum Records in Ethnology. Vol. 1, No. 5: Maori Godsticks collected by the Rev. Richard Taylor. By Dr. T. Barrow. Pp. iii+183 (7 plates). (Wellington, New Zealand: Dominion Museum, 1959) [3011

Chicago Natural History Museum. Fieldiana: Zoology. Vol. 39, No. 32 (September 30, 1959): Two New Game Birds from Peru. By Emmet R. Blake. Pp. 373–376. 10 cents. Fieldiana: Geology. Vol. 10, No. 34 (October 22, 1959) · A Review of the Family Captorhinidæ. By Richard J. Seltin. Pp. 461–509. 1.25 dollars. (Chicago: Chicago Natural History Museum, 1959.) [3011

State of California: Department of Fish and Game. Fish Bulletin No. 107: Trout and Salmon Culture—(Hatchery Methods). By Earl Leitritz. Pp. 169. (Sacramento · Printing Division, Department of Fish and Game, 1959.) 2 dollars. [3011

United States Department of Commerce: National Bureau of Standards. Monograph No. 5: Preservation of Documents by Lamination. By William K. Wilson and B. W. Forshee. Pp ii+20. (Washington, D.C.: Government Printing Office, 1959.) 20 cents. [3011

Machines and Tooling, Vol. 30, Issue No. 5. (Cover-to-cover translation of the Russian technical journal *Stanki i Instrument*.) Pp. 46+8. Published monthly. Annual subscription rates: Ordinary rate within the United Kingdom 70*s*.; Ordinary rate outside the United Kingdom 84*s*. Rate for Libraries of Educational Institutions within the United Kingdom 49*s*.; Rate for Libraries of Educational Institutions outside the United Kingdom 62*s*. (Melton Mowbray · Production Engineering Research Association of Great Britain, 1959) [3011

Conseil Permanent International pour l'Exploration de la Mer, Charlottenlund Slot, Danemark. Rapport et Procès-verbaux des Réunions. Vol. 148: Contributions to Symposium on Salmon and Sea Trout, 1958. Pp. 80. (Copenhague: Andr. Fred. Høst & Fils, 1959) Kr. 16.00. [3011

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THE WANDERING SCHOLAR IN THE BRITISH COMMONWEALTH

A PASSAGE in the Queen's Speech at the opening of the new Parliament last autumn promised legislation to implement recommendations of the Commonwealth Education Conference, and in seconding the address, Mr. E. Gardner not only stressed the importance of seeing that to-morrow's leaders of the Commonwealth have the best and widest education we can give, but also urged the need of a two-way exchange of ideas and learning. He suggested that we should not only welcome Commonwealth students in our universities, our Inns of Court and technical colleges, but should also send our own students to the new universities of Africa and Malaya. In the same debate, Mr. Anthony Head urged much the same from a different point of view, suggesting indeed that it is vital for the West as a whole to overhaul its educational approach to the underdeveloped countries.

Two Bills have already been introduced during the new session which deal with the general problem. The first of these, the Marshall Scholarships Bill, which received its second reading in the House of Commons on November 6 and in the House of Lords on November 26, is essentially a Bill which doubles the number of Marshall Scholarships coming to Great Britain. In moving the second reading, the Joint Under-Secretary of State for Foreign Affairs, Mr. R. Allan, said that during the six years the scheme has been in force, 72 American scholars, including 20 women, had been selected from 37 different colleges and universities in the United States, of whom nine had come from Harvard, six from Stanford, California, and six from Princeton, New Jersey. Twenty-five of the scholars went to Oxford, twenty to Cambridge and fifteen to London, the remainder being spread over eight other British universities; the Marshall Aid Commission would like to see the scholars distributed among more British universities but felt obliged to offer them the university of their choice if vacancies existed there. All in all, the scholarships had achieved the success that had been hoped for, and many suggestions had been made to increase the number of scholars.

The Bill received a warm welcome in both Houses. In the Commons, Mr. P. Noel-Baker associated himself with the tribute paid to the work of the selection commissions in the United States and hoped that these commissions would continue to choose their scholars by regions. Selection simply on academic standards might lead to far too many of the scholarships going to the great universities of the eastern coast of the United States. Mr. P. Goodhart thought that at least one scholarship every year might with advantage go to a graduate of one of the American Service academies, or to the Virginia Military Institute, where General Marshall himself had studied. Mr. M. Stewart referred to the value of such interchanges in interpreting one country to another, and for that reason, he said, it is desirable that the Marshall scholars should be drawn, so far as possible, from every aspect of American life and that they should be given a very wide experience of Britain. It is right that we should respect the scholar's right of choice, but all the more important that we should build up both the performance and the prestige of the newer universities.

Other speakers were just as strongly in support of the scheme, but were anxious to improve it along these lines. Mr. M. MacPherson, for example, wondered whether it would be better to select one student from each State over a period of two years, and suggested that instead of universities in Britain as a group offering places to the twenty-four Marshall scholars, each university should be asked individually to offer a place in a particular subject which it wished to suggest. This might assist better use of some of the special facilities of particular universities, and also correct the concentration on too few subjects. At present students are mainly in social and political science, physics, chemistry and mathematics; of sixty students in the first five years, only two were taking applied science.

While these specific suggestions found little support in the debate, it was generally recognized as desirable to spread the students around so far as possible; and referring to the small number of Marshall scholars studying science, Mr. Allan, replying on the debate, pointed out that the National Science Foundation provides thirty-eight fellowships for Americans to study science. He also promised to put the points raised personally before the chairman of the Commission. In the House of Lords, the Minister of State for Colonial Affairs, the Earl of Perth, who moved the second reading, said that if experience showed that a greater number of scholars than twenty-four was desirable, the Bill would permit such an increase by Order in Council. Indeed, here even more than in the House of Commons, the debate stressed the value of the wandering scholar and the contribution to world understanding which the free intercourse of scholars could offer.

The second Bill, the Commonwealth Scholarships Bill, which received its second reading in the House of Commons on November 25, is the first of the measures promised in the Queen's Speech to give effect to some of the recommendations of the Commonwealth Education Conference. Specifically, it gives effect to the Commonwealth Scholarship and Fellowship scheme described in the White Paper and is intended to introduce a greater measure of academic mobility throughout the Commonwealth at a time when the men and women concerned are at the formative period of their professional careers. As Mr. C. J. M. Alport, Minister of State, Commonwealth

Relations Office, said in moving the second reading, it should provide a continuous exchange of ideas and knowledge by giving them access to the rich and varied intellectual resources and technical experience of the different Commonwealth countries; and he hoped it would go far to create in a very real and literal sense a Commonwealth of the mind, which, applied to the social, economic and political problems of our time, might provide answers to questions which those in the world of public affairs sometimes found so elusive.

The Scholarship and Fellowship Plan is outlined in the report of the Commonwealth Education Conference and the proposed arrangements for the administration of the scheme in the United Kingdom are described in a subsequent White Paper*. The plan is additional to, and distinct from, any other plan already in operation and is based on mutual co-operation and the sharing of educational experience among all countries of the Commonwealth. It is sufficiently flexible to take account of the diverse and changing needs of the countries of the Commonwealth and, while Commonwealth-wide, is operated on the basis of a series of bilateral arrangements to allow for the necessary flexibility. The awards are intended to be designed to recognize and promote the highest standards of intellectual achievement.

If the plan is to achieve its purpose, we must secure the widest possible variety of cultural exchange between all parts of the Commonwealth and so facilitate the development of a multilateral trade in ideas. The Commonwealth Scholarships will represent a system of awards to men and women from all Commonwealth countries who are of high intellectual promise and may also be expected to make a significant contribution to life in their own countries on their return from overseas. In the main, these awards will be made for a period of post-graduate study or research, to men and women capable of reaching the highest standard of intellectual achievement, and the ages of those considered for awards may vary according to the circumstances of particular countries. A limited number of Commonwealth Visiting Fellowships will also be made to senior scholars of established reputation and achievement.

The main emphasis will be on awards in the academic field; but the possibility is not excluded of some awards to other persons who play important parts in the life of their community. Because the awards will be made to persons of high intellectual attainment and held in institutions of higher learning, such institutions and members of the academic community who work in them should play an important part in selecting and placing the scholars and fellows. In the first place, the United Kingdom Government has undertaken to be responsible for the award of a total of 500 scholarships and fellowships to be held at any one time in United Kingdom universities, colleges of technology or other appropriate institutions, and Mr. C. J. M. Alport hoped that the first 250 of these would be available to be taken up at the beginning of the academic year in the autumn of 1960. The Bill creates a Commonwealth Scholarship Commission which will be solely responsible for selecting and placing the Scholars and Fellows. The Commission, of not less than ten or more than fifteen members, at least four of whom will be leading figures in the academic world, including a chairman designated jointly by the Secretary of State for Commonwealth Relations and the Secretary of the Colonies, in accordance with whose directions the Commission will act, will be assisted by a panel of forty advisers consisting of distinguished United Kingdom scholars and scientists. The Association of Universities of the British Commonwealth will provide the secretariat of the Commission, and the British Council will be responsible for paying the cost of passages, fees and allowances and for all arrangements for reception and welfare. When the plan is in full operation, it is expected that the 500 scholarships will involve annual expenditure of about £500,000, with some additional expenditure to supplement awards offered to United Kingdom citizens by other countries of the Commonwealth.

Mr. Alport added that each Commonwealth country is setting up a similar body with broadly the same duties as the Commission in Britain. That Commission will be solely responsible for selecting individuals for scholarships here or recommending candidates for scholarships overseas. He estimated the financial cost at £2,300,000 during the first five years, and urged that in future a period of service or study in the Commonwealth should be regarded not as a handicap, but as an essential qualification for a successful career in the professions, industry or technology in the United Kingdom. He also emphasized the importance of ensuring that students coming to Britain had an opportunity of entering fully into the ordinary life of the country, especially during vacations, and said that special attention would be paid to this by the British Council.

The Bill was warmly welcomed on all sides of both Houses of Parliament, and the debates were marked by much constructive criticism. Mr. H. A. Marquand stressed its value for the exchange of ideas and the exploration of ideas to free the human spirit. The importance of a wide distribution of students using the universities of the United Kingdom was stressed, and the Under-Secretary of State for Commonwealth Relations, Mr. R. Thompson, assured the House that it is intended that the Scholars should go to colleges of technology and not solely to universities. In the House of Lords debate on December 8, the Earl of Home, replying for the Government, anticipated that ultimately some 50 per cent of the Scholars might be studying technical subjects.

The need for special care in the reception of the Scholars was generally appreciated, and in a constructive speech Mr. Geoffrey Lloyd referred to the need to strengthen administrative arrangements in Britain to further the scheme. He suggested also that educational attachés might be useful and that we should consider their appointment to the offices of British High Commissioners and especially those in the younger Commonwealth countries. We must seek also to create an entirely new climate of opinion upon the whole question of Commonwealth education, and Mr. P. Goodhart regarded the Bill as a challenge to the people of Great Britain. The Scholarships would be a direct benefit both to Britain and to the Commonwealth as a whole only if the Scholars and Fellows were not allowed to feel lonely, unwanted and without friends.

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* Commonwealth Relations Office Colonial Office, Commonwealth Scholarship and Fellowship Plan: Proposed Arrangements for the Administration of the Plan in the United Kingdom. Pp. 5. (Cmnd. 894.) (London: H.M. Stationery Office, 1959.) 6d. net.

In replying on this debate, Mr. R. Thompson remarked that the Government would make money available to remove some of the financial obstacles to the recruitment of teachers for the overseas service, and also said that the United Kingdom Government is planning that an additional 500 places in teacher-training establishments of all kinds should be made available for Commonwealth students over the next five years. There was some comment in both Houses on the absence of the Union of South Africa, which has so far not decided to participate in the scheme. Canada is to provide 500 scholarships and fellowships; Australia, 100; India, 100; Pakistan, 30; New Zealand, 25; the Federation of Malaya, 12; Ghana, 10; the Federation of Rhodesia and Nyasaland, 10; Ceylon, 6; and East Africa, 4. Mr. Marquand raised the point particularly in respect of the High Commission Territories in Africa, and thought that now that the universities of South Africa are closed to students from such territories their needs should be specially considered. Dr. H. King referred specifically to the provision by the students of the University of Southampton of a scholarship deliberately aimed at bringing to England some of the students whom the South African Government had denied proper opportunities of education.

At the Committee stage of the Bill in the House of Commons on December 3, comparatively few fresh points were raised: the importance of wide distribution of the Scholars, while giving due consideration to individual preferences, was again stressed; and, for the Government, Mr. C. J. M. Alport emphasized the importance of spreading these Fellows and Scholars as widely as possible over the academic life of the United Kingdom, both geographically and from the point of view of the subjects they came to study. The question of South Africa was considered rather more fully in the House of Lords debate on December 8. The Earl of Home, in introducing the Bill, added little to what had been said in the House of Commons, but stressed the need to ensure that Scholars or Fellows did not suffer in their careers through going abroad for a couple of years in their training. The question of South Africa, however, was raised in forthright manner by the Earl of Lucan. While we would welcome South African participation in the scheme and also South African students in Britain, he did not think it would be consistent with the conception of the Commonwealth on which the Education Conference was based to agree to British students going to South African universities which had been forced to abandon their old liberal traditions and conform to the present racial policy of the Union.

The Commonwealth, it is maintained in the report of the Conference, is a new experiment in human relationships, founded on a belief in the worth and dignity of the individual and a recognition of the value of freedom and co-operative action. Lord Brand, however, argued that it would not be an offence against human dignity to permit a Scholar from Britain to go to the self-governing University of the Witwatersrand, although the University's protests against the policy of the Union Government had proved ineffective. The Earl of Home, however, pointed out that if South Africa decided to come in and offered scholarships in the South African universities, the decision to apply for such scholarships would rest with the students themselves.

Nevertheless, as a side-issue of the debates, the policy of the South African Government towards the universities stood out in the same stark isolation to that of the remaining Commonwealth countries as it had done at the Conference on "Freedom and Responsibility: the Role of the Scholar in Society" arranged by the Committee on Science and Freedom, of which the report has just been published*. This was apparent in Dr. J. W. Cook's introductory paper on university traditions with particular reference to the British Commonwealth, in which he emphasized the importance of academic autonomy and academic freedom, of avoiding any steps which would hinder the free exchange of information, maintaining high academic standards and avoiding becoming involved in political controversy; and also in Dr. F. G. Malherbe's paper on university problems in a multicultural society, which showed factually how in South Africa the Government policy is depriving millions of non-whites of any chance of a university education. Dr. Malherbe pointed out that so far the academic freedom and autonomy of the existing universities continues to stand high; but a note on dismissals of staff at Fort Hare University College circulated with the report raises grave doubts as to how far this can continue.

The debates in Parliament on both these Bills show that Parliament generally is well aware of their importance and of the value of the wandering scholar in the life of the Commonwealth. It is realized, too, that such interchange can only be fully effective if supported by widespread co-operation both on the part of employers to see that the returning Scholars and Fellows are not affected adversely in their careers and equally on the part of the general public in the receiving countries to ensure that, especially in their leisure hours, they are absorbed freely and warmly into the life of the community. Perhaps too little was said as to the value of interchange, not merely in both directions between the United Kingdom and other Commonwealth countries, but also between the Commonwealth countries themselves. Nevertheless, neither the White Paper itself, the Conference report nor the debates in Parliament can leave anyone in doubt as to the value of these imaginative schemes in promoting understanding and goodwill and increasing the supply of trained people, or in developing in responsible men and women the qualities of judgment and wisdom, insight and sympathy, which the advance of civilization increasingly demands.

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* Committee on Science and Freedom. Bulletin No. 13: The Scholar and Society—Report on a Study Group held in Tunis, April 1959. Pp. 144. (Manchester: Committee on Science and Freedom, 518 Wilmslow Road, 1959.)

RIVER STUDIES IN NIGERIA

River Studies and Recommendations on Improvement of Niger and Benue

By Nedeco, Netherlands Engineering Consultants, The Hague. Pp. ix+1000. (Amsterdam: North-Holland Publishing Company, 1959.) 140*s*.

ACCESS from the sea to the Benin ports of Burutu, Sapele and Warri in Nigeria has given much concern ever since about 1930. These ports constitute the link with the inland navigation of the extensive Niger and Benue river system, which provides a waterway of more than a thousand miles into the continent of Africa. At one time ships drawing up to 20 ft. could cross the bar of the Forcados estuary but in more recent years this estuary, along with the Escravos estuary, has only been able to accommodate ships drawing 12–14 ft. A continual decline in the draught available led the Nigerian Government to call in the Netherlands engineering consultant firm of Nedeco to undertake the Western Niger Delta investigation of 1952–54.

Soon after the Western Delta investigation was started preparations were made for a study of the possibility of improving the shipping conditions on the Niger and Benue Rivers. This eventually led the Nigerian Government to request from Nedeco a full-scale investigation of the Niger and Benue river system with the object of determining how the shipping conditions on these rivers could be effectively improved. The consultants were charged with discovering the characteristic nature and behaviour of the component parts of the river system; the composition by means of aerial and other surveys of a series of river maps covering the rivers downstream from Baro and Guara; of ascertaining the régime of the rivers and the vagaries of channel configuration and depths; and inquiring into the feasibility of possible future projects.

The work was commenced in 1954 and Nedeco has now issued a monumental tome of more than a thousand pages illustrated by several hundred diagrams and maps giving the results of the investigations of a team of twelve expert engineers who were stationed in Nigeria for three years. At first glance the report might be thought to be of interest only to those concerned with the practical problems of river navigation, but closer examination reveals that it will be of great value to many other experts. For the consultants found themselves obliged to consider matters which were not directly concerned with the technical problems of river regulation and river management. Since the Niger and Benue systems drain the greater part of Nigeria, the report effectively becomes an up-to-date account of the geography of Nigeria in which the physiography, geology, climatology and hydrology are all systematically considered as a background to the economic traffic on the rivers. It is these that are largely the life-giving arteries of the country. Of the various sections the hydrological naturally receives the most detailed treatment as this was, of course, a prime charge on the consultants. The report concludes with an account of the present problems of navigation, defects of navigability and recommendations for their improvement.

Because the report is so comprehensive it will be found of interest to geographers, geologists, geomorphologists, climatologists, hydrologists and hydraulic engineers: it brings together a great mass of new information and incorporates many new ideas. There can be few of these experts who will not find something of immediate interest in the report's thousand pages. The whole publication is beautifully produced and illustrated, and Nedeco is indeed to be congratulated on the appearance of such an impressive report.

The Western Niger Delta investigation during 1952–54 dealt with access to the ports of Burutu, Sapele and Warri and therefore only that part of the Niger Delta was studied and surveyed which lies west of the Bomadi Creek. Having undertaken the main Niger–Benue investigation it was only natural that the Government began to think in terms of a hydrological survey and investigation of the remaining part of the Delta. Accordingly, an agreement was signed in March 1958 between the representative of the Government of the Federation of Nigeria and of Nedeco for an Eastern Niger Delta investigation. Field-work for this is now under way, and the final report is expected towards the end of 1960.

W. G. V. BALCHIN

SOLUTIONS OF ELECTROLYTES

The Physical Chemistry of Electrolytic Solutions

By Prof. Herbert S. Harned and Prof. Benton B. Owen. Third edition. (American Chemical Society Monograph Series, No. 137.) Pp. xxxii+803. (New York: Reinhold Publishing Corporation; London: Chapman and Hall, Ltd., 1958.) 160*s*.

Electrolyte Solutions

The Measurement and Interpretation of Conductance, Chemical Potential and Diffusion in Solutions of Simple Electrolytes. By Prof. R. A. Robinson and Prof. R. H. Stokes. Second edition. Pp. xv+559. (London: Butterworths Scientific Publications; New York: Academic Press, Inc., 1959.) 65*s*.

The Structure of Electrolytic Solutions

Edited by Walter J. Hamer. (Sponsored by the Electrochemical Society, Inc., New York, N.Y., and the National Science Foundation, Washington, D.C.) (Electrochemical Society Series.) Pp. xii+441. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1959.) 148*s*. net.

OF the three volumes under review, Harned and Owen's "Physical Chemistry of Electrolytic Solutions" and Robinson and Stokes's "Electrolyte Solutions" are new editions of well-known works that first appeared in 1943 and 1955, respectively. "The Structure of Electrolytic Solutions" is a collection of papers given at an Electrochemical Society symposium in Washington in 1957.

Unlike the second edition, which was largely a reprint of the first, the new edition by Harned and Owen incorporates some major revision and has been considerably extended. A section on irreversible thermodynamics has been added, and recent theoretical work on inter-ionic forces and their effects on both equilibrium and transport properties is included. Diffusion of electrolytes is fully discussed. The title "The Physical Chemistry of Electrolytic Solutions" is a pardonable exaggeration; many physico-chemical properties of such solutions are ignored. For example, there is little or nothing on reaction kinetics or spectroscopic properties and, although the adsorption of ions at the air/water interface is discussed, there is no mention of electrocapillarity or electrode reactions. The book is primarily concerned with the thermodynamics and transport properties of solutions and the central theme is the relation of such properties to ionic interaction. Broader and less tractable questions, such as standard free-energies and entropies of hydration, or the limiting mobilities of ions and their relation to ion-solvent interaction, receive less attention. Experimental data are fully discussed and tabulated, but experimental methods are only indicated in outline. In its own field this is a far more comprehensive monograph than any other, and much of its contents cannot be found elsewhere except in the original papers. The treatment is nearly always clear and precise, and the amount of detail given is very nicely judged, being neither too much nor too little. The earlier editions have become accepted as a standard and indispensable work of reference, and I do not doubt that the third edition will continue this tradition.

"Electrolytic Solutions", by Robinson and Stokes, is about half as long as Harned and Owen's book, and costs about two-fifths as much. It may be regarded as an advanced text-book (it certainly serves this purpose well), though in some respects it is rather more specialized in character than a text-book is expected to be. It covers a considerable part of Harned and Owen's territory, but generally in much less detail—with the important exception of experimental methods, which are here described fairly fully. In addition, more attention is paid to the interpretation of the properties of ions in terms of their interaction with the solvent. Profs. R. A. Robinson and R. H. Stokes have deliberately restricted themselves to the discussion of properties that seem to them to be fundamental, and as the sub-title indicates, the main topics are the measurement and interpretation of conductance (including transport numbers), chemical potential and diffusion. Thus, the book is by no means a text-book of general electrochemistry, and even in the narrower field of thermodynamics of electrolyte solutions, it does not deal with heats of dilution. But within their self-imposed limits, the authors have given a stimulating and well-balanced account of the subject, and their book can be recommended without hesitation. The first edition contained extensive tables of experimental results; these have now been enlarged and brought up to date and form a most useful collection of data. Elsewhere some minor changes have been made; for example, recent work on conductance in non-aqueous solvents has been incorporated.

The title "The Structure of Electrolytic Solutions" provides a wide umbrella, under which some twenty-seven papers have found shelter. They cover a big range, both in subject and in character. Some are essentially reviews of work with which their authors have been associated, others give new experimental results and a few are theoretical contributions. A few titles chosen at random include "Proton Transfer in Aqueous Solution", "Ionic Vibration Potentials", "Soret Coefficients", "Conductance and pH in Methanol–Water Mixtures", "Complex Ions in Molten Salts". It is a useful collection of papers and gives the reader a very fair picture of current trends in this field.

All three books are well produced and none suffers from an undue number of misprints or obvious errors. The price of Harned and Owen is formidable, but it is good value for money. J. N. Agar

TROPICAL MARINE MOLLUSCS FROM AMERICA

Sea Shells of Tropical West America

Marine Mollusks from Lower California to Colombia. By Prof. A. Myra Keen. Pp. viii+624+10 plates. (Stanford, Calif.: Stanford University Press; London: Oxford University Press, 1958.) 100*s.* net.

THE wealth of illustrated handbooks on American sea-shells available for coasts of the United States (and less expectedly for other areas) is in sad contrast to the dearth of European manuals. Though several faunal series provide identifications, by means of keys and descriptions, of animals in the North and Baltic Seas, no fully illustrated and reasonably comprehensive handbook on the sea-shells of any European coast has appeared for twenty-five years. For a British book, it is necessary to go back twice that time. Let us hope that the recently mooted European Malacological Union may stimulate the production of a work to challenge the best American models.

Among these, Prof. Keen's book stands high. Within well-chosen limits (excluding only shell-less forms, squid and cuttlefish, minute gastropods, stragglers and deep-water forms) she deals with even the rarest and the most dubious Panamic species, and illustrates all but a few. The coloured plates are decorative, while much of the value of the book lies in nearly seventeen hundred monochrome figures, of generally high quality. These are grouped on uniform pale grey backgrounds into half-tone plates, set in the text so that the illustration of a species occurs on the same pair of facing pages as the concise diagnostic notes. A few explanations of the figures, relegated to an appendix, would be helpful in the text. Though relatively small in bulk, the general matter is well conceived, touching on the ecology, affinities and exploration of the Panamic malacofauna, referring to general texts, and discussing the higher taxa. Ecological information on individual species is inevitably scanty, but the book itself will stimulate the observations by which the deficiency will be rectified.

In such a work, intended mainly for the knowledgeable amateur and the general zoologist, the precise systematic arrangement adopted is of secondary importance; but broader genera might have been adopted in a few highly split groups. Also, subgeneric names would tend less to emphasize difference at the expense of likeness, if merely listed at the head of the contained species, instead of being combined in every species name. Indeed, a slight over-emphasis on nomenclature is the most serious charge that can be levelled against Prof. Keen—whose identification of taxonomy with nomenclature will be ill-received by taxonomists and etymologists alike. That misleading technical term 'type' occurs repeatedly in the addenda though not in the glossary, but will not entrap those who read the suggested works on taxonomic practice. However, the discussion on the name *Mitra sulcata* is comprehensible only, and with difficulty, in the light of a new provision on the status of junior primary homonyms, which many will be surprised to find in the forthcoming edition of the International Rules of Zoological Nomenclature.

The addenda, in fact, are intended not for the general reader but for the systematic malacologist; who should acquire three additional pages, issued separately. They reinforce a bibliography of nearly six hundred titles, with every original reference to an included species, and a large number of original figures reproduced from inaccessible works, to make the handbook invaluable, especially to the isolated worker. Relatively few additional titles would be needed to provide references to the genera also, and publications of one author in one year could be distinguished in the text by means of the letters used in the bibliography. Perhaps these minor improvements may be incorporated in the successive editions which will certainly and deservedly be demanded.

I. C. J. GALBRAITH

BACTERIOPHAGES

Bacteriophages

By Mark H. Adams. With chapters by E. S. Anderson, J. S. Gots, F. Jacob and E.-L. Wollman. Pp. xviii+592. (New York: Interscience Publishers, Inc.; London: Interscience Publishers, Ltd., 1959.) 115*s*.

BACTERIOPHAGES are viruses made up of genetically active deoxyribonucleic acid enclosed within a protein shell, which seem to have no enzymes of their own, with the exception of some forming part of the specialized apparatus, the phage tail, concerned with their attachment to and penetration of the host. Yet the entry of the phage nucleic acid into a susceptible bacterium results in a complete change-over in the metabolism of the host, with synthesis of new enzymes, proteins and nucleic acids.

The advantages of such a system for the investigation of the mechanisms by which specific biosyntheses are controlled and performed are evident. Furthermore, phages, from the technical point of view, are much more convenient to work with than any of the other known classes of virus. It is therefore not surprising that in recent years the phage-infected bacterium has become a favourite experimental material for fundamental research in cell physiology, biochemistry and, perhaps most important, in physiological genetics, and that our knowledge of phages has increased enormously. Phage has been used as experimental material in such a variety of fields that the reports are scattered to an exceptional degree. The first text-book on phage to appear in English for many years has therefore been eagerly awaited. The work in question was begun some years ago by Dr. Mark Adams, of New York University College of Medicine, well known for his own researches on phage, particularly in the neglected field of their taxonomy, and for the course which he taught at Cold Spring Harbor for many summers; a course which served as an introduction to phage for many biologists and workers from other fields who have since contributed largely to our increasing knowledge of this subject.

At the time of Mark Adams's untimely death in 1956, his manuscript was unfinished. It has been completed by the addition of four chapters, planned but not drafted by Adams: on "Chemical Interference with Phage Growth", by J. S. Gots; on "Lysogeny" and on "Colicins", by F. Jacob and E.-L. Wollman; and on "Use of Phages in Epidemiological Studies", by E. S. Anderson. The eighteen chapters finished or partly written by Adams have been revised, brought up to date and completed by a panel of distinguished workers in phage research, the whole work being under the general editorship of A. D. Hershey.

The book as thus completed forms an excellent and reasonably detailed account of our knowledge of phage. The style and presentation are satisfactory, and even those without previous knowledge of the subject should have no difficulty in following the argument. Indeed, it may perhaps be said that at some points simplification has been carried too far; for example, the equation derived by Visconti and Delbrück for the frequency of a recombinant class among the progeny phage released after infection of bacteria by a mixture of phages of differing genotypes is given, but not its derivation. However, the qualitative treatment given will probably suffice for most readers. Those requiring more detail will find themselves well served by the extensive bibliography (some 900 references); it is to be regretted that not all the citations include the titles of papers. A glossary of terms used in the literature is also included in the volume.

Despite the rapid progress of research on phage, most experiments even now make use of a rather limited number of more or less standard techniques. One of Mark Adams's most useful services to the phage world was the publication in 1950 of a review entitled "Methods of Study of Bacterial Viruses", in which all these indispensable experiments and techniques were described and discussed. This most valuable compendium has been included as an appendix, with such additions and amendments as were needed to bring it up to date.

All the principal aspects of phage are dealt with, and it is satisfactory to be able to record that the earlier literature, mainly European, has not been neglected. The use of phages in epidemiology is the only application of phage described; their use in the treatment of bacterial infections, to which so much attention was given in the years immediately following the discovery of phage, is scarcely mentioned, an omission perhaps justified by the disappointing or equivocal results obtained, both in experimental and natural infections. In general, it may be said that the selection of results from the extensive literature has been admirably made, for while nearly every report of general importance is discussed or mentioned, the very large number of papers of only ephemeral or specialized interest are not referred to.

This work can therefore be warmly recommended (and indeed its high price seems to me its only major fault). It will certainly become, and long remain, the standard source of information on phage. It forms a worthy memorial to its principal author, both as scientist and teacher.

B. STOCKER

ELECTRONIC SECTOR-SCANNING ASDIC: AN IMPROVED FISH-LOCATOR AND NAVIGATIONAL INSTRUMENT

By Prof. D. G. TUCKER and Dr. V. G. WELSBY

Electrical Engineering Department, University of Birmingham

UNDERWATER acoustic echo-ranging systems are used extensively[1] in the form of vertical-beam echo-sounders for navigational and surveying purposes and for fish-location, and in the form of horizontal-beam asdics for fish-location and (especially during the Second World War) for the detection of submarines. In these systems considerable success is achieved in the simpler objectives of determining the depth of a level sea-bottom beneath a ship, of determining the depth of a fish shoal provided it happens to be under the ship, and—specially in the case of horizontal-beam systems—even in searching for fish-shoals. But the performance of all these existing systems is severely restricted by the fact that they are single-beam systems. A new system which we and our colleagues have developed retains most of the characteristics of the existing devices, but gives simultaneous information over a wide sector of search with good angular and range resolution. It therefore greatly aids the use of the echo-ranging system for searching, and speeds up and improves the accuracy of survey work. Moreover, it can eliminate the need for mechanical movement of the often heavy electro-acoustic transducer and thus greatly reduce the cost of ship-fitting operations.

In the existing single-beam systems the pulse is transmitted by a transducer (or loudspeaker), and received, after reflexion from the sea-bottom or whale or other object, by the same or a different transducer (this time used as a microphone). The time difference between transmitted and received pulses measures the range of the bottom or object, and is recorded on a chart by a stylus moving at a speed equal to the velocity of sound in water multiplied by the ratio of the width of the chart to twice the maximum range of detection. The transmission of the pulse marks the left-hand edge of the paper, and any echo received causes a mark at a point in the stylus sweep corresponding to the range of the object causing the echo. The paper is meanwhile moving at right angles to the stylus sweep, so that the next pulse transmission marks on a line adjacent to the previous one. The repetition of the echo in the same (or almost the same) position on successive sweeps causes a very great increase in the detectability of the echo if it is a weak one, and if the ship carrying the equipment is moving, the profile of the sea-bottom, or the shape of any object in the beam, is slowly traced out. To look in different directions means rotating the transducer(s), and while such a facility is usually provided in horizontal-beam asdics, it is not usually provided in echo-sounders.

The new system has the following distinctive features: (*a*) The receiving acoustic beam is relatively narrow and is swung continuously over a large sector by electronic means; the rate of sweep is so high that the sector is covered within a time less than the duration of the transmitted pulse. Thus, if the transmitting beam covers the whole sector, then echoes arising anywhere in the sector are received during each pulse repetition interval. (*b*) The received information is displayed on a cathode-ray tube in which one axis of deflexion corresponds to bearing (by being synchronized to the beam-swinging) and the other to range. The intensity of the spot indicates the strength of the echo.

This electronic sector-scanning system clearly enables the shape of the bottom profile over a certain length, or of any object in the sector, to be displayed instantly. The advantages of this in fish-location (and in identification of the fish) and in navigation and surveying may be considerable, and may be judged to some extent from the photographs of actual displays which accompany this article.

Outline of System

A schematic diagram of the system is shown in Fig. 1. The receiving transducer is n times the length of the transmitting transducer, and is divided into n sections, where n is the number of beam-widths (as measured between points where the power response

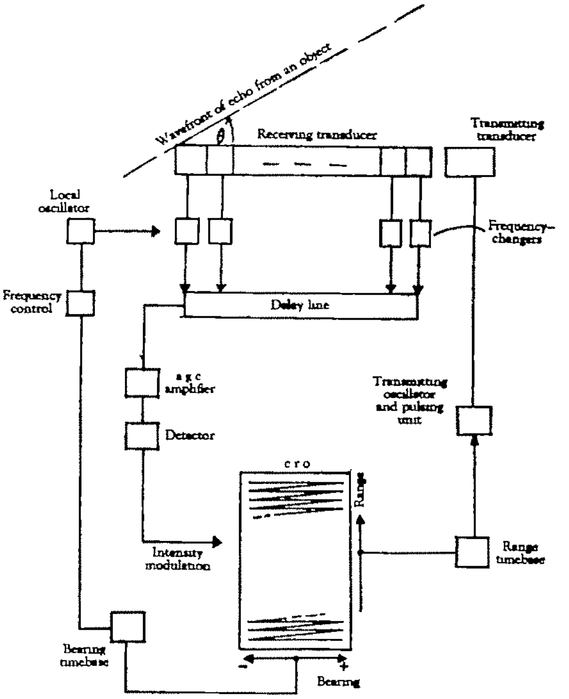

Fig. 1. Schematic diagram of electronic sector scanning system

has fallen to half) it is desired to contain in the scanned sector. If these *n* sections were connected to *n* corresponding, uniformly spaced, taps on the delay line, then the beam would be deflected by an amount dependent on the phase-shift in the delay line. The principle of this is readily seen: if an echo is received from a target on a bearing of θ relative to the perpendicular axis of the transducer, then the wave-front of the echo pulse lies parallel to the line shown in Fig. 1. The echo is received by the first transducer section before it reaches the second, and so on. Thus the echo signal received by successive sections is delayed in phase relative to the previous sections. If a peak output is to be obtained for this particular angle of arrival, then the delay line is required to insert compensatory phase-shift so that all the components of the combined output are in the same phase.

In the scanning system, frequency-changer equipment is inserted between the transducer sections and the delay line. The local oscillator which feeds all the frequency-changers is swept in frequency by the bearing time-base, so that the signal frequency received by the delay line varies over a range during every sweep of the bearing time-base. If then the delay line is made to have a phase-shift which varies over this frequency-range from negative values to positive values, the beam will be swept from left to right during each sweep of the bearing time-base. The latter also deflects the spot on the cathode-ray tube from left to right, so that signals received on any particular bearing are recorded on that bearing on the display. The range time-base works in the usual way, so that the position of an echo-spot on the tube indicates the position of the echoing object on rectangular axes of bearing and range. If the bearing scan is so rapid that it is completed within the duration of the pulse and is immediately repeated, no information is lost, and all directions in the sector are effectively looked at simultaneously, but with the angular resolution corresponding to the beam-width of the receiving transducer.

The technical and design considerations of this system have already been published elsewhere[2,3].

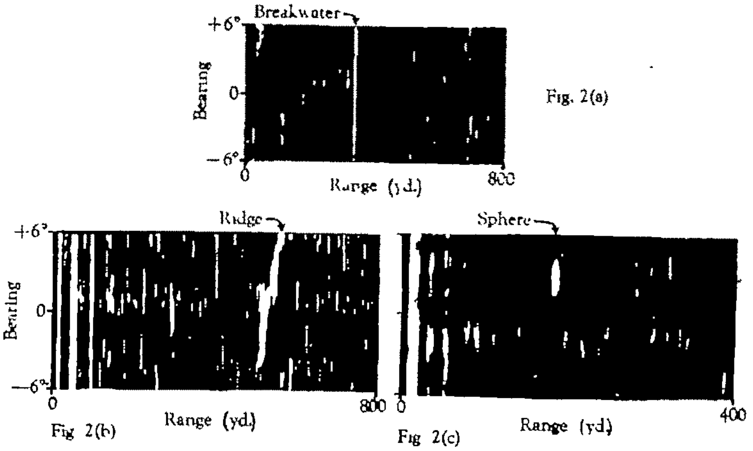

Fig. 2(a)
Fig 2(b)
Fig 2(c)

Fig. 2. Some photographs of sector displays taken on R.R.S. *Discovery II*. Beam nearly horizontal (tilt 5° for *a*, *b* and *c*). *a*, Portland Harbour, eastern breakwater viewed exactly at right angles. (N.B. The target echo was so strong that the brilliance was turned down and the echoes from the sea-bottom are lost.) *b*, Ridge on sea-bottom near the Dorset coast. *c*, 18-in. diam. sphere in mid-water at range of 150 yd. in Weymouth Bay. (Several successive traces have been superposed in the photograph as the sphere was moved out by a small boat.) 400-yd. range scale

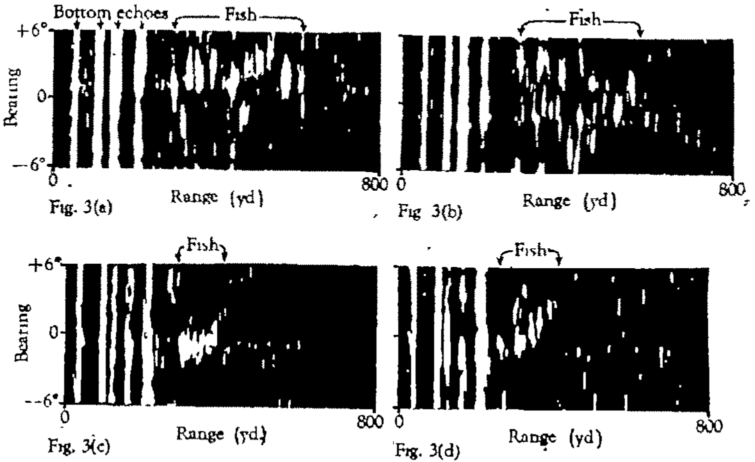

Fig. 3(a)
Fig 3(b)
Fig. 3(c)
Fig. 3(d)

Fig. 3, *a*, *b*, *c* and *d*. Small fish shoals in southern North Sea. (800 yd range scale)

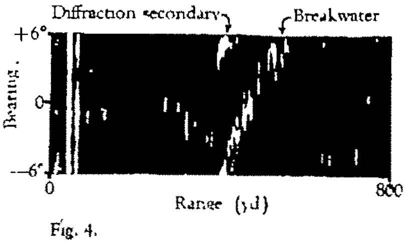

Fig. 4.

Fig. 4. Photograph of full sector display of outer breakwater of Portland Harbour viewed from the east; showing effect of 'side-lobe interference' due to a 'diffraction secondary'

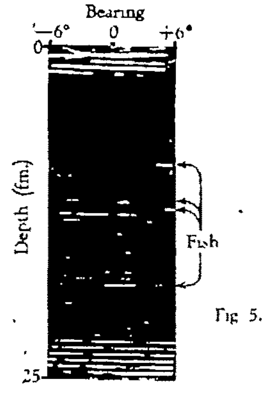

Fig 5.

Fig. 5. Scanning echo-sounder display with fish passing through sector. (Southern North Sea, depth about 20 fathoms, max. range on scale = 25 fathoms)

Results obtained with Nearly Horizontal Beam

The first laboratory experimental equipment, made in the University of Birmingham, was, through the courtesy and co-operation of the National Institute of Oceanography, given sea-trials in R.R.S. *Discovery II* during October 1958 and November 1959. On the latter occasion the equipment was used as a research tool by the staff of the Fisheries Laboratory, Lowestoft, in a study of fish migration. The ship was already equipped with a suitable electro-acoustic transducer designed and constructed by the National Institute of Oceanography. The primary object of the equipment and the trials was the location and study of fish shoals—some small fish shoals are shown on the display in Fig. 3*a*, *b*, *c* and *d*—but photographs of the way in which objects of navigational and surveying interest appear on the display were also obtained. A selection of these latter is shown in Fig. 2*a*, *b* and *c*. Fish can be distinguished from fixed objects, of course, by their movement and the variation of the shape of the shoals. For example, the shoal in Fig. 3*d* is the same as that in Fig 3*c*, but observed a minute or two later. The general speckled background in the photographs is the reverberation, or general back-scattering of sound from the sea-bed and waves. This, being random, never repeats from one pulse to another, and genuine targets can be easily distinguished, not only by their brightness, but also by their consistency from pulse to pulse.

The parameters of the equipment were as follows :

Beam-width = 1·5° in horizontal plane (between ½-power points)
Scanned sector = 12° in horizontal plane, that is, 8 beam-widths
Vertical beam-width = 10°
Pulse duration = 1 m.sec.
Frequency = 37 kc./s.
Transmitted acoustic power = 100 watts during pulse

A technical description of the equipment has been published elsewhere[3].

It should be noted that the heavy lines across the display in the range 0–100 yards approximately are due to the various side-lobes of the vertical beam-pattern striking the sea-bottom at ranges depending on their angle relative to the main beam. In general, no object on the bottom can be reliably detected until after the last of these lines. In an operational system the side-lobes of the vertical beam-pattern would probably be largely removed by suitable transducer design, and the early part of the range, up to the point where the main beam reaches the bottom, would be left clear for the detection of objects in mid-water. But in the experimental trials, the transducer was simultaneously being used for other experimental work which required the side-lobes to be present. The position of the beam in the vertical plane was, however, kept constant by a stabilizing mechanism using a gyro as reference, so that the angle of tilt of the main axis (quoted in Fig. 2) is a known constant for any series of observations.

The fact that there are only 8 beam-widths in the scanned sector in the experimental equipment accounts for the fact that even a 'point' object, such as the 18-in.-diam. sphere, extends over at least one-eighth of the bearing scan. An operational system might with advantage scan more than 20 or more beam-widths.

It is shown in a previous paper[2] how, at the edges of the scan, a 'target' just moving off at one edge begins to show at the other, but as movement proceeds further, the false echo extends with ever-decreasing strength. This is not serious for most purposes, but can be avoided by sacrificing some of the scan and placing a mask over the edges of the display. An illustration of the effect is shown in Fig. 4. It may be possible to get better results by more complex transducer arrangements, and this is being investigated. The effect is in some ways similar to 'side-lobe interference' in radar, but is more correctly known as 'diffraction secondary interference'.

The maximum useful range of detection is of the order 800–1,000 yards at the frequency chosen for the experimental equipment.

Use with Vertical Beam

When the transducer is tilted so that the beam points vertically downwards, the equipment becomes a scanning echo-sounder. If the beam is wide from fore to aft, but narrow from side to side, and scans from side to side, then the display will show, for each pulse transmission, the sea-bottom profile in some detail across the track of the ship. The ship's own motion forwards enables the profile along the track to be recorded. Thus *one* traverse of an area enables a three-dimensional picture of the bottom to be obtained—a result which shows an enormous saving of cost and time in surveying an area. It is, of course, the slant range which is shown on the display, and a correction must be made for true depth.

The vertical-beam operation is also useful for fish location and identification. Fig. 5 shows a photograph of the display obtained when a very scattered shoal of fish (probably herring) was passing through the sector. Individual fish are probably indicated by the smallest of the more distinct marks.

Conclusions

The results obtained by the biologists studying the movements of the fish will be published separately. It can be said, however, that the equipment has already proved itself able to give information on the horizontal and vertical distribution of the fish, and more particularly on their movements, which has probably not been obtained before ; and it may well prove an important research instrument in fisheries and hydrographic work. A system with much higher resolution in range and bearing is being developed, and it may eventually prove possible to study the movements of individual fish, of nets and other objects ; the need already exists.

To summarize, it is thought that the new electronic sector-scanning asdic has considerable possibilities in : (*a*) detection and location of fish ; (*b*) identification of fish by their shoaling habits, echo-strength and shape, etc. ; (*c*) research in fisheries work, for example, in studying the structure and internal and overall movements of fish shoals, and even of individual fish ; (*d*) navigation ; (*e*) hydrographic surveying.

We are grateful to our colleagues in the Electrical Engineering Department at the University of Birmingham for much of the research, design and constructional work which has been involved, and to the National Institute of Oceanography for some financial assistance, for help with the trials, and for the use of R.R.S. *Discovery II* and of the transducer. The interest taken in the work, the use made of the equipment, and the further financial help given by the Fisheries Laboratory, Lowestoft, together with much encouragement and interest on the part of the Marine Laboratories, Aberdeen, have proved a great stimulus.

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[1] Tucker, D. G., *J. Brit. Inst. Rad. Eng.*, 16, 243 (1956).

[2] Tucker, D. G., Welsby, V. G., and Kendell, R., *J. Brit. Inst. Rad. Eng.*, 18, 465 (1958).

[3] Tucker, D. G., Welsby, V. G., Kay, L., Tucker, M. J., Stubbs, A. R., and Henderson, J. G., *J. Brit. Inst. Rad. Eng.*, 19, 681 (1959).

THE HARWELL NEUTRON PROJECT

By DR. M. J. POOLE and DR. E. R. RAE

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A LABORATORY has been set up at the Atomic Energy Research Establishment, Harwell[1], to exploit the possibility of using a linear electron accelerator as a pulsed neutron-source for time-of-flight studies. These studies increase our knowledge of highly excited states of compound nuclei.

The first machine to be used in this way produced electrons of energy 3 MeV., and was superseded in 1952 by a 15-MeV. machine, which produced photo-disintegration and photo-fission neutrons from a block of uranium which served as target for the electrons. In that accelerator the duration of the electron pulses could be made as short as 0·1 μsec. by pulsing the electron gun, and the beam current during a pulse was of the order of 25 m.amp. at 13·5 MeV. under full-load conditions (400 pulses/sec.). In these circumstances the rate of production of neutrons during the pulse was 10^{14}–10^{15} neutrons/sec.

The new laboratory has been designed around a much more powerful machine than either of its forerunners, since it is capable of providing a beam current during the pulse of the order of 400 m.amp. at 30 MeV. Using this in conjunction with a natural uranium target, a rate of emission of 2×10^{16} neutrons/sec. is possible. However, a further increase in the available neutron output has been obtained by building a special target (called the neutron booster) which is a multiplying sub-critical uranium-235 assembly, and by this means a ten-fold increase in the neutron output has been obtained. Thus the final rate of production of neutrons during the pulse is 3×10^{17} neutrons/sec. (A factor of 1·5 is accounted for by the difference in the production-rate of primary neutrons in natural uranium and uranium-235.)

In the time-of-flight experiments the neutrons are partly moderated in flat water tanks, 2 cm. thick, placed on three sides of the neutron booster. A layer of boron-10 metal placed between these tanks and the uranium-235 core prevents return of slow neutrons to the uranium-235 and so avoids serious distortion of the pulse shape. Some nine windows lead through the shielding and allow neutrons to pass down evacuated 'flight tubes' arranged radially about the neutron source and so reach the experimental apparatus. In this way the time-of-flight, and so the energy of the neutrons associated with some nuclear process, can be measured by recording the time-interval between the accelerator pulse and the time of arrival of the neutron. The recording of the times of arrival of the neutrons is carried out by multi-channel time analysers, one of which is associated with each individual flight path. The ability to carry out a number of time-of-flight experiments simultaneously in this way is one of the main advantages of a pulsed accelerator over a fast chopper as a pulsed neutron-source. The other advantage is the relative ease with which neutron pulses as short as 0·25 μsec. are obtainable.

Experiments in progress include measurements of neutron partial cross-sections for scattering, capture and fission, in addition to total cross-sections. From these measurements, the parameters of nuclear levels extending for several tens of keV. above the neutron binding energy are extracted. These parameters include the level-spacings, the neutron, radiation and fission widths, and the angular momenta. Another type of experiment is the study of the hard γ-rays arising from the capture of a neutron into various levels of the compound nucleus above the binding energy. An analysis of the relative strengths of these energetic γ-rays in different levels in the same compound nucleus demonstrates the random fluctuations that occur in the strength of a particular γ-ray transition.

The new machine should make possible a much more thorough investigation of the statistical laws governing the distributions of the parameters of nuclear levels, and may help to clear up certain mysteries which still surround the phenomenon of nuclear fission. In addition to these studies of neutron interactions, the investigation of a variety of photo-nuclear phenomena is also planned.

In the field of reactor physics, the pulsed-source method has been extended to allow the measurement of neutron spectra in reactor lattices. A knowledge of these spectra is of great importance in calculating the detailed performance of reactors, particularly the phenomena which govern the amount of permissible burn-up. Calculation is in principle possible once a detailed knowledge of the neutron-scattering law is known ; but neither experimental results nor methods of calculation are yet sufficiently developed to make this feasible, and recourse has to be made to direct experiment. In this method the target of the pulsed machine is placed inside a section of the lattice of interest, and the neutrons are moderated down to the spectrum characteristic of that lattice. This spectrum is then measured by the usual time-of-flight method.

The increased intensity from the new accelerator will make possible the performance of more complex experiments in which a pulse of neutrons is introduced into a moderator and the spectrum measured by using a synchronized neutron shutter (chopper) which opens a known time after the electron pulse. In this way it will be possible to follow the change in neutron spectrum during the last stages of slowing down, and to make a direct comparison with some of the theories of neutron thermalization available. It will also make possible the direct observation of the so-called 'neutron cooling', that is, the reduction in mean energy of neutrons in a moderator due to preferential escape of fast neutrons.

[1] Poole, M. J., Wiblin, E. R., *et al.*, Proc. Second Int. Conf. on Peaceful Uses of Atomic Energy, Geneva, 1958 (Paper P/59).

FORESTRY COMMISSION RESEARCH STATION

BEFORE 1946 the Forestry Commission had no research station; a limited amount of work on silviculture and mensuration was carried out by Forestry Commission staff at headquarters and elsewhere, and work on some other subjects, such as entomology and pathology, was done by specialists attached to other institutions.

In the summer of 1946, Alice Holt Lodge, a moderate-sized country house attached to an old Crown forest, was released from its war-time military occupation and opened as a Forest Research Station. By 1950 this building was proving inadequate for the expanding needs of the Station, and many of the offices were moved into huts. It was obvious that this could only be a temporary measure, and plans for more permanent expansion culminated in the spring of 1959, when a new wing, considerably larger than the building to which it is attached, was ready for occupation. This three-storey wing, together with a single-storey service block and a staff restaurant, has a total floor area of nearly 25,000 sq. ft. Construction is in the form of a concrete framework, supported on piles sunk 36 ft. into the Gault clay. The outer walls are made up of prefabricated panels, containing large timber-framed windows, with timber cladding below. The internal corridors are off-centre, giving rooms 16 ft. deep, suitable for the larger laboratories, on the eastern side, and rooms 12 ft. deep for offices and personal laboratories on the western side. The architects were Messrs. Cowper and Poole, of London.

The two buildings now serve as headquarters for all the research work of the Forestry Commission, with the exception of that on machinery and on utilization development, which is based on London. A branch station in Edinburgh, controlled from Alice Holt, deals with silviculture, genetics and to some extent management problems in Scotland and the north of England. However, much of the work centred on Alice Holt extends into Scotland. Although a silvicultural nursery, a tree-breeding nursery, several seed-orchards for the production of improved strains of tree seed, two poplar nurseries, a botanical collection of poplars and a pathological test nursery are attached to the Station, Alice Holt forest is not itself a research forest, and the experiments on which the staff of the Research Station are engaged are in forests and nurseries scattered all over England, Scotland and Wales.

Much of the field work is supervised by out-stationed research foresters, but nevertheless considerable travelling on the part of the Alice Holt and Edinburgh staff is also involved. In research nurseries or at centres where there is a large number of forest experiments, the Research Branch has its own labour forces, but much of the often considerable work involved in laying down forest experiments is carried out with the aid of the ordinary labour force of the forest concerned. It has been found that the progress and success of the work depend very considerably on the maintenance of a proper balance between the graduate staff planning and analysing experiments and the research foresters responsible for their lay-out, supervision and assessment in the field. There is no doubt that the existence of this body of skilled foresters provides a vital link in forest research, which is lacking, or at any rate insufficient, in some other forest research organizations even in countries where more money is available.

The new wing at Alice Holt houses those research sections which have need of laboratory space. Silviculture, with its subsidiary Sections, Ecology and Soils, occupies the whole of the top floor. Silviculture is concerned with all aspects of the cultivation of forest crops from the seed stage onwards. In the past, emphasis in Britain has inevitably been on nursery problems and on the establishment of plantations; but, as Forestry Commission plantings get older, work is becoming more concerned with matters such as the thinning, pruning and manuring of established plantations. Attention is now being given to methods of regenerating felled woodlands. This poses a number of difficult problems that will become more acute when earlier Commission plantings reach maturity. Felling will, of course, provide opportunities for change of species or variety, so that trials of different species, and in particular of different seed origins within those species, are still being actively pursued.

The Ecology Sub-section is concerned with studies of the site relationships of individual tree species, and with the investigation of sites where the establishment of satisfactory tree crops has proved difficult or impossible. The Soils Sub-section has two overlapping functions, analytical services for other sections and in particular for silviculture, and a programme of research into the nutrition of forest trees. Work has been particularly concentrated on foliar analysis and this is proving an essential part of the proper understanding of the many experiments on forest manuring now in progress.

On the second floor, but still under the control of the Silviculture Section, is the seed laboratory. This has a programme of work on improvements in seed-testing technique and on the storage of seed, but its main activity is the testing of all seed lots bought by the Forestry Commission, and for this purpose it is licensed as a 'private seed testing station'. Much of the seed which it tests is housed in the central Forestry Commission seed store, which occupies one end of the ground-floor of the new wing. This is not a research responsibility, but is housed at Alice Holt for convenience, and so that advice on storage and testing facilities are readily available. The store has three large refrigerated chambers, two for routine storage at 2° C. and one at −5° C. for the more difficult species. The total capacity of these chambers is 42,500 lb. of conifer seed. All the conifer seed bought from abroad or collected in England and Wales is stored at Alice Holt.

One end of the second floor is occupied by the Genetics Section. This could be more correctly described as a 'Tree Breeding' Section, since it is concerned mainly with practical problems on the improvement of forest trees. The selection of 'plus' trees from existing stands has already led to the formation of seed-orchards of Scots pine, Douglas fir, European larch and a few other species. Hybridization, followed by further selection, is being used to effect still further improvements.

The remainder of the second floor is occupied by the Pathology Section, which is concerned with the study and control of tree diseases. The work includes not only damage by fungi and bacteria, but also that due to non-living agencies, such as frost. At the moment, the control of *Fomes annosus*, the most serious cause of butt rot in conifers in Britain, is the major project of this Section. Other projects are top-dying of Norway spruces, a physiological disorder associated with increased exposure of the tree crown, and the testing of poplar and elm clones for resistance to bacterial canker and elm disease, respectively.

The Entomology Section, on the ground-floor, is engaged on investigations of a number of forest pests. It is normally concerned with the biology, including the occurrence of parasites, the forest status, which usually involves population sampling, and the control of each insect. In some cases, for example that of the pine looper moth (*Bupalus piniarius*), work has been particularly directed to finding the population-level at which the insect becomes a potential danger. In another case, that of the larch sawfly, work is directed to finding the effects of different populations and, therefore, of different degrees of defoliation, on tree growth.

The remainder of the ground-floor is occupied by the Photographic Section, which provides a common service to the rest of the Station, as well as to the Forestry Commission as a whole, taking most of the photographs required for illustration and record, and the slides necessary for lecturing purposes. The collection of more than 12,000 black-and-white photographs and nearly 7,000 colour slides is arranged numerically, but fully card-indexed on the International Forest Decimal Classification, so that photographs or slides on any particular subject can easily be located. The Section has produced several short films on forestry subjects.

The old building houses Administration, the Management Section, the Statistics Section, the Library, and a large room, which is used for meetings, conferences, lectures, etc.

The Management Section includes mensuration, which studies the growth of individual trees and of crops, and the methods of measuring them. It produces volume tables, yield tables, etc., for use in forest management. Management also includes the Census of Woodlands, and therefore has charge of all the data resulting from the census of all woodlands of 5 acres or more, carried out between 1947 and 1949, and of the census of hedgerow and park timber carried out in 1951. Working from this basis, sampling surveys for general purposes and local surveys for special purposes will be used in the future to provide more up-to-date information. Now that the hitherto overriding need for a strategic reserve of timber has become less acute, it is obviously desirable that more attention should be given to the economic background of forestry in Britain. For this reason, the Management Section now includes an economist, who is studying the nature and volume of the demand for timber and relating these to the costs involved in raising it. The Management Section is also responsible for assisting the Forestry Commission field officers in the preparation of forest working plans.

The Statistics Section is again a common service to the whole Station. The statistician advises on the layout of all experiments, thus ensuring that the resultant data are suitable for computation. The long-term nature of, and the large areas involved in, many forest experiments make care over design of particular importance. Computation is now done on a 'Pegasus' electronic computer by arrangement with the Royal Aircraft Establishment at Farnborough. The Section also carries out research into the application of statistical methods to problems of forest research and management.

The Library is a small one, containing some 3,500 books; but it receives all the principal forestry periodicals and has a considerable collection of reprints, reports, etc. It also carries out documentation work, resulting in a large card index of literature references on British forestry and on aspects of forestry abroad which are of interest to British foresters. There is also a series of classified information files. All filing is arranged according to the International Forest Decimal Classification.

The Forestry Commission encourages more fundamental work on some of its problems by means of grants to universities and other institutions. This has enabled the Alice Holt Research Station to concentrate the efforts of its staff very largely on the more immediately practical aspects of forest research. Thus, despite the relatively small staff—36 research officers and 107 research foresters in the Research Branch as a whole—there is a considerable output of work, mostly published in Forestry Commission publications (*Forest Records*, *Bulletins*, etc.) or in forestry and other scientific journals. A summary of the work of the Station, with brief accounts of grant-aided work at other institutions and with a number of short papers by the Research Station staff, is published annually as a Forestry Commission Report on Forest Research.

OBITUARIES

Prof. J. G. Wood

THE sudden death of Prof. J. G. Wood on December 8 last at the comparatively early age of fifty-nine has not only deprived the University of Adelaide of its senior professor but also has been a severe loss to biological work in Australia.

Joseph Garnett Wood was born at Mitcham, South Australia, on September 2, 1900, and educated at the University of Adelaide, where he obtained his first degree in the honours school of chemistry (1922). Fortunately he had shown interest and ability in botany during his undergraduate course, and in 1923 he became a demonstrator in botany. A programme of work on the vegetation of arid South Australia was beginning and the assistance of a chemist was vital. The saltbush (*Atriplex vesicaria*) which covers huge plains in the arid parts of the State had been termed a halophyte on insufficient evidence. Wood collaborated in the ecological work, which showed that the soils were not saline, except for the scattered salinas where *Atriplex* was absent. The plant had a high content of sodium chloride in the cells of its uncuticularized, tomentose leaves. Wood investigated the transpiration-rates of a number of trees and shrubs growing in the region and later showed that, in addition to having a low transpiration-rate, *Atriplex* was able to take up moisture through its leaves from saturated air. This work led to the award of an 1851 Exhibition scholarship, and Wood proceeded to Cambridge (Caius College) to read for a Ph.D. On his return to Adelaide in 1927 he was appointed lecturer, soon to be in charge of the Department. He was promoted to professor in 1935.

Work on the vegetation of arid South Australia had led to the generous donation to the University of an area of some 1,000 acres and small field laboratory at Koonamore, about 200 miles north of Adelaide in 1926. Wood collaborated in the work of the Koonamore Vegetation Reserve almost from the start. It is a tribute to his abiding interest in the arid flora that the Reserve and laboratory are still in regular use by the Adelaide Department for teaching and research.

Wood's contributions to botany fall under the heads of general ecology (his work on "The Vegetation of South Australia" (1937) has long been out of print), autecological work on arid plants and nutritional studies. Recently (1957) he and one of his research students have shown that sodium chloride is an essential micronutrient for *Atriplex vesicaria*. The nutritional studies have dealt with nitrogen and sulphur metabolism and the role of such micronutrients as copper, zinc and molybdenum. These are essential for crop production over large areas of leached soils in Australia but are not needed in the same quantities by the native sclerophyllous plants.

Wood had considerable organizing abilities. The fine range of biological buildings at the University owes much to his planning. Possessed of an alert mind and known for his fairness and even temper, much of his time and energy of late years have been spent on committee work. He was a member of the Interim Council of the Australian National University during 1948–51 and a Council member during 1952–55. He served on the Advisory Committee of the Commonwealth Scientific and Industrial Research Organization during 1951–59 and was chairman of the Board of Standards, of the Australian Journals of Scientific Research, and chairman of the Board of Research Studies of his University at the time of his death.

His recreations were largely connected with his home and garden. He had a good knowledge of Australian contemporary art and he himself painted. In 1930 he married Joan Hazel, who, with their three daughters, survives him. His one-time teacher and colleague can only join his many other friends in deploring his sudden passing.

T. G. B. Osborn

Prof. W. R. Fearon

William Robert Fearon, professor of biochemistry in the University of Dublin, died on December 27, 1959, aged sixty-seven years. The only child of a Presbyterian clergyman who had died when his son was four years old, he entered Trinity College, Dublin, in 1911. It was an indication of the intellectual versatility which was to characterize his life that he found some difficulty in deciding whether to read English or science. In the event he read natural science, in which he graduated B.A. with a Senior Moderatorship in 1915, securing also a Foundation Scholarship in Experimental Science two years later. He entered the Medical School during this period, but became so intensely interested in physiology and biochemistry that he interrupted his medical studies in the fourth year to do research work in nutrition in London during the First World War. Following this, he was awarded an exhibition to Emmanuel College, Cambridge, where he became one of the devoted band which Frederick Gowland Hopkins gathered around himself in the University Biochemical Laboratory. On his return to Trinity College, Dublin, in 1921, he was elected to a fellowship and thus began a period of almost forty years during which he enhanced the reputation of the College with his researches, delighted his students with the brilliance of his lecturing and entertained his colleagues by his sparkling conversation on almost any topic.

His literary bent had found recognition while he was still a student in the award of the vice-chancellor's prize in English verse, the University Philosophical Society's Gold Medal for English composition and the editorship of the College miscellany, *T.C.D.*, to which he was a frequent contributor. Throughout his career his clear, limpid style was to stamp everything he wrote whether scientific or otherwise. He was a master of the happy descriptive phrase which left an indelible impression. Always interested in motor-cars, he wrote of a visit to the Lancia showrooms in London during the early '30's that he had been taken for a drive in a car "that was the size of a cottage and cost as much as a house".

His early work was concerned with the formation of urea from protein, in the course of which he began to exhibit that facility for developing colour reactions which was to lead to so many papers in the *Biochemical Journal*. He loved to demonstrate these reactions to his colleagues or to visitors, and he must have discovered far more than he ever published. His interest in nutrition persisted and found its most recent expression in a survey of the fluoride content of Irish drinking waters. It was typical of his alert mind that he began this work more than ten years ago, at a time when others were only commencing vaguely to realize the possible practical significance of fluoride intake in dental health. For about six years before his death he had been interested in the ureides obtained from autolysed yeast. An outstanding contribution was his book, "An Introduction to Biochemistry", first published in 1934; he was working on the fourth edition when he last became ill. It is remarkable in that it is at once "An Introduction" which unfolds the subject to the student systematically and with the utmost clarity, and a comprehensive review that can be consulted with profit by the more mature worker.

Notwithstanding a rather shy disposition, he had wide interests outside his laboratory and, indeed, outside College. In the '20's he was the friend and associate of the literary and artistic giants of the time in Dublin—W. B. Yeats, Jack Yeats, George Russell (AE), Lennox Robinson, Oliver St. John Gogarty. He held the honorary professorship of chemistry of the Royal Hibernian Academy. His play, "Parnell of Avondale", was a success when produced at the Abbey Theatre nearly twenty years ago. He was an accomplished pianist and organist, and had an extensive knowledge of music.

One of the fascinations of knowing Fearon was the frequency with which, even after years of close friendship, one discovered new interests of his: a large stamp collection, to which he added regularly; his flair for cars, both vintage and modern sports—his own collection had included a Lancia, a Bentley and a Rolls; his real appreciation of the musical comedies of the early part of this century. In the same way, one learned with surprise, in 1943, that he was a candidate for a seat in the Senate of the Irish Parliament as a representative of the University of Dublin, for one had never been aware of his interest in matters political. Yet he continued to hold his seat to the end, and his colleagues in the Upper Chamber often expressed a high regard for him as a member.

William Fearon was a man of great gentleness of character but one who did not encourage intimacy, and not many of his friends knew him really well. Nevertheless, his passing will leave many gaps. He will be missed in the College Common Room and in his clubs as in the class-room and in the Senate chamber.

W. J. E. Jessop

NEWS and VIEWS

Experimental Physics at Liverpool:

Prof. J. M. Cassels, F.R.S.

Prof. J. M. Cassels, who has retired from the chair of physics in the University of Liverpool, joined the staff as a lecturer in 1952. In 1955 he was promoted to senior lecturer and in 1956 was appointed to the professorship of experimental physics which had been instituted a few years before but had remained vacant. He had gone to the Atomic Energy Research Establishment as a research fellow in 1949 on completing his Ph.D. at Cambridge, having previously done some war work there. At Harwell, he joined the research group working on the new 170-MeV. synchrocyclotron and played a leading part in the experimental programme on *p–p* and *n–p* interactions. He transferred to Liverpool to work with the higher energy (380 MeV.) Liverpool cyclotron, which came into full operation at the end of 1954. Largely because of Cassels's foresight, they had a full experimental programme ready to go, and this has continued at a steady pace ever since. Cassels ran a small group of his own, and his name is on many of the laboratory's papers, especially some concerned with pion interactions. But his influence went far beyond his own group, and inspired much of the high-energy work of the laboratory. His strength is both in experimental techniques and in his grasp of theory. He is also an enthusiastic teacher, and a very fine lecturer. He resigned his post to take up, in the autumn of 1959, a visiting professorship in Cornell University, where he will be able to continue his work on high-energy nuclear physics. In 1959 he was elected a Fellow of the Royal Society.

Dr. A. W. Merrison

Dr. Merrison, who has been appointed as Prof. Cassels's successor, was educated at King's College, London, and after taking a degree went to work on centimetric radar at the Signals Research Development Establishment. In 1946 he transferred to the Atomic Energy Research Establishment, Harwell, and worked on various aspects of neutron spectroscopy in the Nuclear Physics Division. He was appointed as lecturer in the University of Liverpool in 1951 and transferred to the research appointment known as the Leverhulme lectureship in 1954. He became a group leader on work with the large 380 MeV. synchrocyclotron and published several important papers on pion physics. In the spring of 1957 he was appointed to a staff post at CERN, the international high-energy laboratory at Geneva. There he worked with the 680-MeV. synchrocyclotron which started work shortly after. One of the results of his team, the discovery of the rare mode of decay of a pion directly into an electron instead of through the normal muon, is considered as of exceptional importance in confirming the theorists' general conception of these particles. Recently, Merrison has been concerned with arranging work for the 28-GeV. proton synchrotron at CERN which has just started working. Dr. Merrison had no opportunity of taking a normal Ph.D. course in a university on account of the War, and so took his degree as a member of the University of Liverpool staff. He is expected to return to the University in the autumn of this year.

Geological Survey and Museum:

Sir William Pugh, O.B.E., F.R.S.

Sir William Pugh, director of the Geological Survey of Great Britain and the Museum of Practical Geology, is retiring at the end of July. Sir William, who was born on July 28, 1892, was educated at the County School, Welshpool, and at the University College of Wales, Aberystwyth. After service in the First World War, in which he was made O.B.E. (Military Division), received the French Croix de Guerre and was twice mentioned in dispatches, he was appointed in 1919 professor of geology at Aberystwyth. In 1931 he was appointed to the chair of geology in the University of Manchester, and became pro-vice-chancellor and later deputy vice-chancellor of that University. He was awarded the Murchison Medal of the Geological Society in 1952 for his researches into the stratigraphy of the Palæozoic rocks in Central Wales. Sir William became director of the Geological Survey and Museum in 1950. He was elected a Fellow of the Royal Society in 1951, and knighted in 1956.

Dr. Cyril J. Stubblefield, F.R.S.

Dr. Cyril J. Stubblefield is to succeed Sir William Pugh as director of the Geological Survey of Great Britain and the Museum of Practical Geology. Dr. Stubblefield was educated at the Perse School, Chelsea Polytechnic and the Royal College of Science, London. In 1923 he was appointed demonstrator in geology in the Imperial College of Science and Technology, a post he occupied until 1928, when he joined the Geological Survey as a geologist. In 1929 he entered the Department of Palæontology of the Survey; and in 1947 he was appointed chief palæontologist. This post he held until 1953, when he was promoted to an assistant directorship in charge of Special Services, and Northern Ireland. Dr. Stubblefield has made many important contributions to our knowledge of the palæontology and stratigraphy of the Palæozoic rocks, and in particular has made a special study of the trilobites. His contributions to geological science have received recognition from the Geological Society of London, by the award of the Bigsby Medal in 1941, and, in 1955, the Murchison Medal. He was elected to the Royal Society in 1944. Dr. Stubblefield's geological activities have not been restricted to his work on the Geological Survey, for he has been an active member of various scientific societies. He is at present president of the Geological Society of London, vice-president of the Palæontographical Society, and a member of the Council of the British Association.

Willis Rodney Whiting Award : Dr. Morris Cohen

Dr. Morris Cohen, of the National Research Council, Ottawa, is to be awarded the Willis Rodney Whiting Award for outstanding work on corrosion. The presentation will be made in Dallas, Texas, on March 14 by the U.S. National Association of Corrosion Engineers. Dr. Cohen is head of the Corrosion Group in the Division of Applied Chemistry, Canadian National Research Council. His work has been largely in physical chemistry, finding what forms corrosion takes, identifying corrosion products, determining rates of corrosion in pure metals and metal alloys. The investigations have included the polishing, etching and electroplating of metal surfaces in order to study the effects of such treatment on the resistance of the metals. Dr. Cohen was born at Regina in 1915, received his B.A. degree at Brandon College in 1934, and his M.A. and Ph.D. degrees at the University of Toronto. He joined the staff of the National Research Council in 1943.

U.S. Navy's Bathyscaphe *Trieste*

On January 24 the U.S. Navy's bathyscaphe *Trieste* successfully dived to a depth of 37,800 ft. in the Pacific Ocean. The region selected for the dive was the Marianas Trench, the depth of which had previously been estimated at 5,364 fathoms (32,184 ft.). Pressure measurements on the bathyscaphe reached 16,883 lb. per sq. in. during the half-hour stay on the ocean floor. The total descent was accomplished in 4 hr. 42 min., and the ascent in 3 hr. 17 min. The operation was directed by M. Jacques Piccard and Lieut. Don Walsh.

New Research Qualification

The Royal Institute of Chemistry has established a Research Diploma for work in pure or applied chemistry to be awarded to graduate members, associates or Fellows of the Institute under conditions similar to those for the Ph.D. degree of a university. The Research Diploma will be particularly directed to those who are unable to satisfy the requirements for taking a higher degree of a university, but who may be able to undertake research under an approved supervisor in a technical college, an industrial firm, a Government department or a research association. The scheme will apply equally to university graduates and those who qualified at technical colleges. Further information can be obtained from the Assistant Registrar, Royal Institute of Chemistry, 30 Russell Square, London, W.C.1.

International Computer Federation

Eleven nations have now ratified the status of the newly-formed International Computer Federation. The Federation was formed following the first international conference on Information Processing, sponsored by Unesco and held in Paris during June 1959. At a provisional committee for the Federation the following members were named : *Chairman*, Isaac L. Auerbach (United States) ; *Vice-chairman*, Prof. A. A. Dorodnicyn (U.S.S.R.) and A. van Wijngaarden (Netherlands) ; and *Secretary*, J. A. Mussard (Unesco). It is expected that the first meeting of the Council of the Federation will take place later this year, resulting in plans for a second international conference on information processing, with an associated technical exhibit in 1963. Representation of the United Kingdom is by the British Computer Society, Ltd., Finsbury Court, Finsbury Pavement, London, E.C.2, from which further information can be obtained.

First All-India Congress of Zoology

At the first All-India Congress of Zoology, sponsored by the Zoological Society of India and held at the University of Jabalpur during October 24–28, the president, Dr. M. L. Roonwal, director of the Zoological Survey of India, referred in his address to the great number of species comprising the fauna of India and pointed out how little is known of the biology of many of even the commoner ones. He also directed attention to the rapidly decreasing numbers of some of the larger animals and appealed for more effort in conservation. Turning to field ecology, he emphasized what a wide field is available for research which can be done with the aid of a microscope and quite simple equipment. He added, however, that what the worker has to create for himself is a suitable mental atmosphere which will enable him to do hard, arduous work, perhaps for years on end, and without thought of rushing into print too soon merely to increase the number of his published papers. Dr. Roonwal concluded with a plea for the training of more systematists to work on the national zoological collections in the charge of the Zoological Survey of India. The Congress which followed was divided into six sections at which more than a hundred papers were presented, and symposia on marine zoology and fishes in the Indian Ocean and on the re-orientation of teaching of zoology in India were held.

The Coryndon Memorial Museum, Nairobi

Despite the fact that Dr. L. S. B. Leakey, curator of the Coryndon Memorial Museum, has been heavily engaged with work on the Olduvai human remains and lectures in Britain and elsewhere, steady progress is recorded in the annual report for 1958. Probably the two most important exhibits during the year were the Shakespeare Second Folio and living examples of rare snakes. The former is placed in a special case and pages are turned periodically. The live snakes form a major attraction and included gaboon viper, rhinoceros horned viper, boomelang, garter snake and the rare Storms' water cobra. Another exhibit was that of the striped weasel which is only the second to be recorded in Kenya. With the giant human teeth from Olduvai and a Chellean I stage of culture, an ivory tool from the Chellean II level and some samples of red-ochre used by hand-axe man are shown. It is interesting to note that a new method of skinning small mammals has been devised, and a paper on the process is in course of preparation.

Palæoliths at Leopoldville

H. van Moorsel, keeper of the prehistoric and geological collections at the Lovanium University at Leopoldville, has published an article in the Faculty of Sciences on the palæolithic finds in the neighbourhood of the city. There is a vast stony plain in the vicinity which seemingly is very ancient, as it does not appear to have any connexion with the existing river terrace systems. Archæological finds have been few in number, but the author claims to have recognized Sangoan and Middle Stone Age tools. It is difficult to judge from reproductions of photographs and possibly some of the suggested tools figured are really naturally fractured, but there is no reason to think that these cultures were not represented. The article was read as a paper at the fifth Congress of Prehistoric and Protohistoric Sciences at Hamburg in 1958.

Central Electricity Generating Board Research Fellowship

The Central Electricity Generating Board is offering a research fellowship, tenable at St. Catherine's College, University of Oxford, in the physical sciences, including metallurgy, engineering or mathematics. The fellowship, tenable for three years, will take effect from October and will be valued at £1,000 per annum. It is open to persons of British nationality, who by the date of his or her appointment have taken the degree of doctor of philosophy or gained the equivalent experience in research. Applications should reach the Master elect, St. Catherine's College Planning Office, 42 Banbury Road, Oxford, not later than March 31.

Refresher Course on Science in Schools

The Department of Extra-Mural Studies and Education of the University of Durham, King's College, Newcastle, is organizing, in conjunction with the North-Eastern Association of the British Federation of University Women, a refresher course on Science in Schools. The course is intended primarily for women with scientific training who, having had to interrupt their careers, now wish to take up or return to teaching in schools. The course will take place at the University during March 23–30. Further information can be obtained from Miss C. Cruse, 292 Ovington Grove, Newcastle upon Tyne, 5.

University News : Belfast

The following appointments are announced : Dr. D. J. Carr (Melbourne), to the chair of botany ; Dr. R. A. Henstock, to be senior lecturer in pure mathematics ; and Dr. A. L. Stewart, to be lecturer in applied mathematics.

Monash

The following first appointments of staff at Monash University, the newly established University in Victoria, Australia, are announced : professor of biology, Dr. A. J. Marshall, reader in zoology and comparative anatomy at St. Bartholomew's Hospital Medical College, London ; and professor of chemistry, Dr. R. D. Brown, senior lecturer in general chemistry, University of Melbourne.

Announcements

Sir Patrick Linstead, rector of the Imperial College of Science and Technology, University of London, has been appointed chairman of the Science Advisory Committee of the British Council and a member of the Executive Committee of the Council in succession to the late Sir Alfred Egerton.

Sir Harold Roxbee Cox, vice-chairman of the National Council for Technological Awards, has been appointed chairman in succession to Lord Hives, who has retired. Sir Harold Roxbee Cox, formerly chief scientist, Ministry of Fuel and Power, is deputy chairman and chairman-elect of the Metal Box Co., Ltd., and a director of Wilmot Breedon (Holdings), Ltd., and of Boulton Paul Aircraft, Ltd. ; he is a member of the Council for Scientific and Industrial Research.

The Scottish Branch of Aslib is organizing its annual general business meeting and a one-day conference to be held on February 5 in the Scottish Central Library, Lawnmarket, Edinburgh. Further information can be obtained from Miss N. I. Shinie, T. and H. Smith, Ltd., Edinburgh 11.

The twelfth international symposium on Crop Protection is to be held at the Agricultural College of the University of Ghent on April 26. The symposium will coincide with the twenty-fourth five-yearly international Floralia of Ghent. Further details can be obtained from Prof. J. van den Brande, State Agricultural College, Ghent, Belgium.

The joint annual meetings of the Chemical Society and the Royal Institute of Chemistry will be held in Belfast during April 5–8 at the David Keir Building of Queen's University, Belfast. Further details can be obtained from the Joint Annual Meetings Officer, Royal Institute of Chemistry, 30 Russell Square, London, W.C.1.

The spring meeting of the Clay Minerals Group of the Mineralogical Society will be held during April 12–13 in the Department of Chemistry at the University of Aberdeen. The topic for discussion will be "Clay Minerals in Sediments". Further details can be obtained from Dr. R. M. S. Perrin, School of Agriculture, University of Cambridge.

Three courses of six lectures each will be given at the Borough Polytechnic, Borough Road, London, S.E.1, on the following topics : Variety Reduction in Engineering (commencing on February 17) ; Lubrication and Lubricants (commencing on February 22) ; Foundry Sands (commencing on February 9). Further information can be obtained from the Secretary of the Polytechnic.

The Association of Oil and Colour Chemists has announced that the twelfth Technical Exhibition will be held at the Royal Horticultural Society's New Hall, Greycoat and Elverton Streets, London, S.W.1, during March 15–17. Further details can be obtained from the secretary, Mr. R. H. Hamblin, Association of Oil and Colour Chemists, Wax Chancellor's Hall, Gresham Hall, London, E.C.2.

Prof. A. Masevich, vice-president of the Astronomical Council, U.S.S.R. Academy of Sciences, Moscow, is to give a number of lectures throughout Great Britain on "Conquest of Space". These lectures are being organized by the Society for Cultural Relations with the U.S.S.R. at the Royal Festival Hall, London, February 6 ; Birmingham Town Hall, February 8 ; Colston Hall, Bristol, February 9 ; and the Free Trade Hall, Manchester, February 12. Tickets are available from the agents of the various halls.

SHELL DEPARTMENT OF CHEMICAL ENGINEERING, CAMBRIDGE

IN March 1945 the University of Cambridge gratefully accepted the offer of the Shell Group of Oil Companies to found a School of Chemical Engineering. This generous gift amounted to £435,400 and was accompanied by an offer to make an annual sum of £2,500 (later increased to £3,000) available until further notice, thereby forming the Shell Chemical Engineering Studies Fund.

The first Shell professor of chemical engineering, T. R. C. Fox, took up his appointment in October 1946 and teaching started in October 1948. Since that time 187 students have graduated in chemical engineering and the teaching staff has grown to its present establishment of six lecturers, three demonstrators and an assistant director of research (two of these posts are vacant at present).

The teaching in the Department consists of a two-year course taken either after Part 1 of the Natural Sciences Tripos or Part 1 of the Mechanical Sciences Tripos. This course leads to the Chemical Engineering Tripos, normally taken in the fourth year. From the outset it has been the aim of the Department to provide a scientifically based training both for those who have been led to the subject through their interest in chemistry and for those whose primary training has been in engineering. It is not the intention to mould these two groups into a single pattern but rather to modify both and let them temper one another. In this way the Department has pioneered a new development in chemical engineering education.

Graduates in chemistry or engineering from other universities may also be admitted to the course, which qualifies them for the B.A. degree.

In the early days the Department was housed in a temporary single-storey building in Tennis Court Road; eventually 9,000 sq. ft. of temporary accommodation was provided on this site. After the exploration of many alternative schemes, a permanent site adjacent to the Cavendish, Metallurgy and Zoology Laboratories was allocated in 1955. Mr. J. Murray Easton was appointed architect and a building with a net floor area of 22,000 sq. ft. was designed to replace and incorporate parts of the old chemical laboratory. Work on the building started in November 1956 and the Department began to move into the first section of its new building early in 1959. The main group of laboratories was opened by Lord Godber, chairman of the Shell Group of Companies, on December 9, 1959 (Fig. 1).

The Laboratories of the Department are partly in a new steel-framed building of five floors and partly in the four-storey building along the Pembroke Street frontage, originally built in 1907, which has been virtually rebuilt internally. The last stage of the rehabilitation of the 1907 building (the ground floor now occupied by the Chemistry Laboratory) will begin in June 1960. The two buildings have been welded into a whole by a new staircase which feeds floors in the converted building from half landings.

The basic design envisaged relatively large-scale experiments being carried out at the lowest level (in what the Department terms the Unit Operations Laboratory (Fig. 2), which is 48 ft. × 45 ft. × 22 ft. high) with adjacent service workshops, stores and instrument laboratory. On the first floor are centred the teaching activities with lecture rooms, library, chemical laboratory and cloakroom. On the second and third floors are eleven combined office-research laboratories for teaching staff, five small research laboratories and one large research laboratory for chemical bench work. Existing structural and service limitations restricted the use of the converted 1907 building. A bank of six constant-temperature rooms (four having 16–17 ft. headroom) are built in the region of the junction between the two buildings, and on the first floor advantage was taken of the headroom to provide a ramped lecture theatre capable of seating both first- and second-year classes together. The basement is used for stores and mechanical services and a new roof structure was built to provide a drawing office and common-room in place of a group of attics at various levels.

In the Unit Operations Laboratory which passes through two floors there are galleries to permit access to apparatus at 4 ft. and 11 ft. above floor-level. There is also a central well 5 ft. deep and provision in five places to pass through the floor of the laboratory above, so increasing the effective ceiling height at these points from 22 ft. to 35 ft.

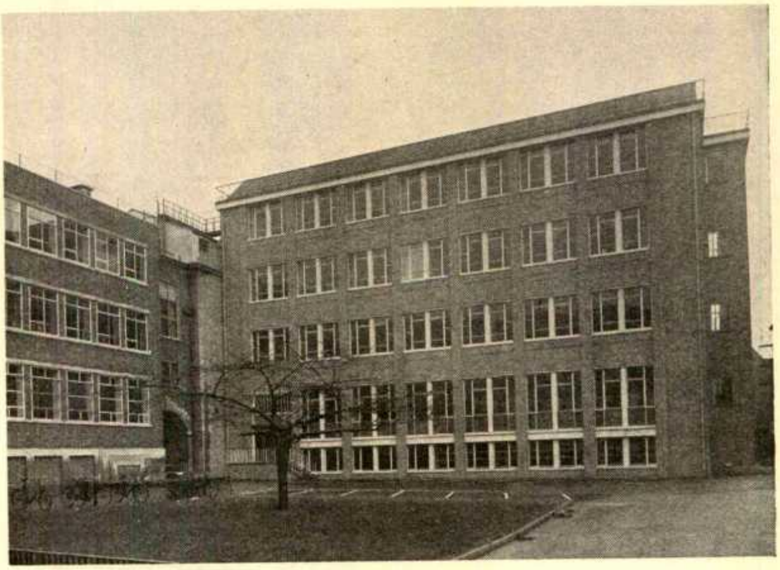

Fig. 1. New building for the Department of Chemical Engineering, Cambridge

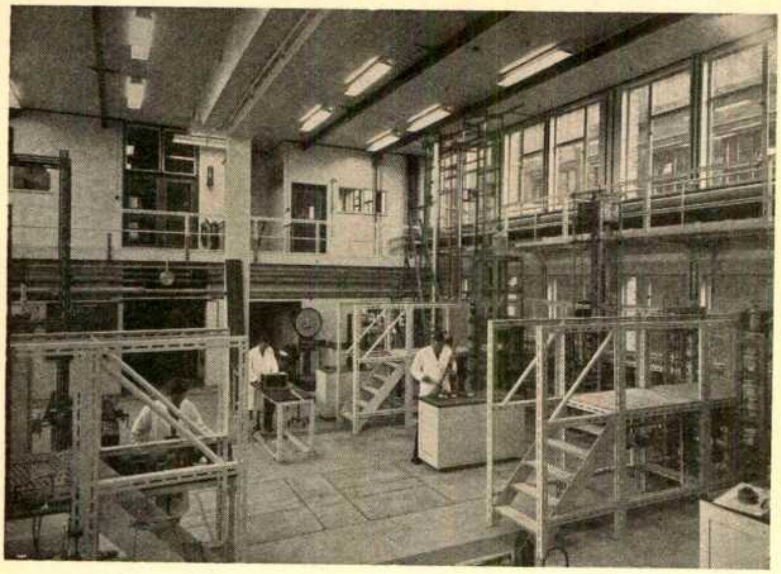

Fig. 2. Unit Operations Laboratory

Each experimental position in the laboratory has steam (10 and 100 lb./sq. in.), compressed air (10 and 100 lb./sq. in.), gas, water and electrical (60 amp.) services. These services are provided by: (1) two steam ring mains operating at variable pressures and supplied by three boilers with a combined capacity of 2,500 lb./hr.; (2) two compressed air ring mains (one 5 in. in diameter for part of its length) operating at variable pressures up to 100 lb./sq. in. and supplied by four reciprocating compressors providing up to 540 c.f.m.; (3) two water ring mains fed from a 6,000 gal. capacity tank in the roof. In the case of both air and steam there is a cross-over pipe network to permit any compressor or boiler to feed either of the pair of ring mains. Additionally there is a general ventilation extract system, balanced by a warmed air intake, and there is also a furnace extract system.

Fume cupboards in the research laboratories have been installed in the centre of the building in spaces between ducts. The fume cupboards are of the 'walk in' type and as the tray at bench height is removable they may be used for high towers and similar apparatus. They are finished internally with an epoxy resin. The cupboards are arranged in vertical banks of three, each bank being exhausted by a single fan. Two of these banks have interconnecting, lined trunking to permit the installation of apparatus approximately 6 in. × 12 in. through three floors, that is to say, approximately 40 ft. high in what becomes effectively a single fume cupboard. Provision has also been made to install apparatus if required in ducts (5 ft. 6 in. × 3 ft. 6 in.) which run almost the full height of the building. These ducts are now fitted with removable floors and used as cupboards.

In the earlier years of the Department's existence, research work necessarily took second place to the development of the teaching programme. However, its importance has steadily increased, and to date twenty-one research students have submitted work for research degrees.

A feature of the Tripos course is that students carry out a modest research project during their final year, rather than a formal laboratory course in chemical engineering operations. Some information of genuine value has been obtained.

The industries or other fields of employment entered by graduates on leaving are as follows:

Petroleum and petrochemicals	29
Chemicals (excluding petrochemicals)	80
Chemical plant manufacture	21
Atomic energy	23
Miscellaneous	34

The original staff of the department is now widely dispersed. Prof. K. G. Denbigh was the first professor of chemical technology at Edinburgh, Prof. E. S. Sellers became the first professor of chemical engineering at Swansea, Prof. J. M. Kay is at the Imperial College, London, and Prof. P. V. Danckwerts has recently returned from the Imperial College to take up the Shell professorship following the resignation of Prof. T. R. C. Fox, who is to continue teaching work in Cambridge in the Engineering Laboratory.

THE SCIENCE MASTERS' ASSOCIATION

NEARLY seven hundred science teachers from many parts of the British Isles gathered for the annual meeting of the Science Masters' Association, which was held in the University of Southampton during December 29, 1959–January 1, 1960.

There was a large attendance at the Guildhall on the evening of December 29, when members of the Association were welcomed to Southampton by the Mayor, Mrs. R. M. Stonehouse. The civic reception was followed by a lecture, illustrated in colour, on "Southampton Borough Archives" by the Borough archivist, Mr. Bruce Jones, and members were able to examine some of the historic treasures of Southampton, which were displayed with models and illustrations of modern educational buildings prepared by the Borough architect, Mr. L. Berger.

The programme included lectures and demonstrations, visits, discussions, films and, in addition to the usual members', manufacturers' and publishers' exhibitions, a number of special exhibitions which attracted considerable interest.

The morning sessions were devoted to lectures and discussions which fully maintained the high standards of the annual meeting. Of special interest to physicists were the lectures by Prof. A. M. Taylor on "Some Demonstrations in Light and Colour" (which filled the large physics lecture theatre to overflowing on two successive mornings), Mr. S. Weintroub on "The Solid State", Mr. B. H. Venning on "Analogue Computers and their Use as a Teaching Aid", and Mr. E. Wolfendale on "The Junction Transistor and its Application". Chemists enjoyed lectures by Dr. G. W. A. Fowles on "Directed Valency" and Dr. H. M. Frey on "Reactions of 'Hot' Molecules", while the interests of biologists were stimulated by lectures by Prof. W. T. Williams on "The Applications of the Pegasus Digital Computer to the Solution of Problems in Plant Ecology and Taxonomy", by Dr. K. A. Munday on "Modern Developments in the Use of Cooling in Physiological and Biochemical Studies", and by Mr. T. H. Blank on "Research on Game Birds". Many teachers found pleasure and profit in lectures not immediately connected with their own branches of science, notably in hearing Prof. E. J. Richards on "Some New Problems in Aeronautical Research", Dr. N. S. Grassam on "Nuclear Engineering—Past, Present and Future", and Dr. K. Hecht on "Atomic Physics in West German Schools".

A most interesting programme was arranged for afternoon visits to industrial establishments, various departments of the University, and places of general interest. The traditional exhibitions were well supported; the sixty manufacturing firms represented had put on a fine display and reported exceptionally keen interest in their products, while there was a steady stream of members to see the excellent publishers' stands in the pleasant setting of the Lanchester Engineering Building. The members' exhibition, always a popular focal point at the annual meeting, and housed this year in three laboratories in the Department of Physics, showed all the usual fertile ingenuity and craftsmanship that is an object lesson and a stimulus to those who are not so skilful or so inventive.

An important feature of the meeting was the special exhibition on "Atomic and Molecular Structure and Nuclear Energy", with some fine displays on this theme by firms and other organizations in the United Kingdom, the United States and West Germany. These included a student reactor which employs natural uranium fuel with a light-water moderator and a small polonium–beryllium neutron source, a reactor simulator and monitoring equipment, and many pieces of apparatus and teaching aids which would be valuable to schools, technical institutions and universities. Equipment from America included a micro-wave analogue of the Bragg diffraction apparatus, using aluminium balls to simulate the crystal lattice structure, an ingenious α-particle counter, and a neat apparatus for the determination of Planck's constant. There was an impressive display of equipment which is available for the teaching of atomic physics in West German schools. Other special exhibitions included visual aids, a feature display on textile research, and apparatus and teaching aids contributed by teachers in secondary modern schools.

During the meeting simultaneous discussions took place on draft syllabuses in physics, chemistry and biology, prepared by subject panels set up by the Association to implement the policy statement, "Science and Education", which was published in 1957. A fourth panel is working on a syllabus for education in science as part of general studies in the sixth form.

A most enjoyable discussion by the Science and Religion Group of the Association was initiated by Dr. Donald MacKay, of King's College, London, who took as his theme "The Christian Doctrine of Man".

A real highlight of the Southampton meeting was touched on Wednesday evening, when Dr. D. G. James, vice-chancellor of the University, gave his presidential address to a packed audience in the refectory of the University, following the Association dinner and University reception. Dr. James, speaking on the subject of "Literature and Science", emphasized the imperative need to bring the sciences and the humanities together in education; calling for more intellectual excitement and the necessity to look beyond one's own special interests to try to see them in relation to others and to the abiding questions, he concluded: "It cannot be doubted that, as boys and girls in ever-increasing thousands move on to the sixth form and the universities, the enjoyment of literature and the arts by the majority who will read science may not be sacrificed except at a severe cost. At the same time, it is clear that the life of science shares with the arts the power of the creative imagination, and that fellowship of the scientist and the artist, instead of being, if Sir Charles Snow is right, a rare thing, should be the most natural thing in the world".

The marked growth of the Association in membership and in its links with other organizations was a major topic at the business meeting on December 31.

From a membership of 2,700 in 1945, the number had grown to 4,500 in 1955 and is now 6,500, the growth continuing at a steadily increasing rate. This rapid development has thrown an increasing burden on the officers of the Association, and the chairman, Dr. H. F. Boulind, emphasized the urgent need for a permanent headquarters with salaried staff to enable the work of the Association to be carried on effectively.

Regarding the links of the Association with other bodies, it was pointed out that, with the growing realization of the significance of science in education and the consequent activity by many organizations and individuals, there was bound to be some duplication of effort through ignorance of what others were doing, some waste of resources and some inefficiency. This indicated an urgent need for a co-ordination centre as a home for science education. This would at least reduce some of the overlapping and wasteful use of personal and financial resources; more positively, this venture could have a really valuable effect on science education not only in Britain but also more widely, for example, through Unesco and the Organization for European Economic Co-operation, through associations of science teachers in other countries and through individuals using the Centre. The Association already has many valuable links with other organizations both in Britain and abroad; others are being forged, and the meeting was very glad to welcome Prof. H. F. Meiners, of New York, who had come to Southampton to discuss the extension of the happy co-operation between the Association and the American Association of Physics Teachers. The Science Masters' Association is sponsoring the establishment of the proposed Science Education Centre in January 1960 as one way of celebrating the diamond jubilee of its foundation. The Centre is being set up temporarily at Holt, in Norfolk, but it is hoped that sufficient financial and other resources will be put at the disposal of the Association to provide more adequate accommodation and equipment in a less-remote spot in the near future.

H. Lockett

ECOLOGY OF PARASITES

THE autumn meeting of the Parasitology Group (Institute of Biology) was held in London at Birkbeck College on October 30, 1959. In the opening discussion of the meeting, which was entitled "The Ecology of Parasites", Prof. B. G. Peters (Imperial College, London) considered the effect of soil factors on plant parasitic nematodes. Soil-living stages are directly influenced by edaphic factors such as moisture, temperature, chemical constitution of the soil and the presence of predators and of possible hosts. Prof. Peters went on to point out that in the parasitic phases, complex relationships exist between the biomasses of host plants and parasitic eelworms. With increasing plant nutrients in the soil there is an increase in host biomass and a correlated increase in parasite biomass. However, at a fixed nutrient-level, the correlation between host and parasite biomasses is negative and the parasites retard the development of their hosts. In species of *Heterodera* having one generation each season, the multiplication rate is highest at low population-levels. As the parasite population density builds up so the rate of multiplication falls. Concurrently, the host at a fixed population density shows a correlated fall in biomass until it can barely maintain the eelworm population. Under these conditions, catastrophic crop losses occur in one season and can alternate with reasonable crop yields the next.

Dr. R. Lainson (London School of Hygiene and Tropical Medicine) spoke about the adaptations shown by coccidial parasites to their environment. The life-histories of sporozoan Protozoa show striking modifications which may be regarded as allowing the parasite to overcome the hazards of transmission and which form an evolutionary pattern, often serving as a basis of classification.

Eimeria, a typical coccidian, has one host in its life-cycle and the difficulties of transfer are overcome by the production of resistant oocysts, which can remain viable outside the host for long periods. The hæmogregarines have eliminated the dangers of a free-living stage and are passively transferred by ectoparasitic invertebrates such as leeches, ticks and mites. The parasites circulating in the blood cells of the vertebrate are ingested by a blood-feeding invertebrate, which itself is eaten by a new vertebrate host.

Yet other coccidians are further adapted and undergo most of their life-cycle in the ectoparasite, the vertebrate virtually serving as a reservoir of infection for the invertebrate host. Besides these modifications in the mechanism of transmission, changes have taken place in the processes of fertilization and sporogony, and all these adaptations ensure maximum efficiency in the maintenance and dissemination of the species.

Dr. S. D. Garrett (Botany School, Cambridge) discussed inoculum potential. Infectivity denotes the potentiality for infection of a single infective organism. In order to produce disease in 50 per cent of inoculated host individuals a large dose of infective agents is often required, and the term 'inoculum potential' denotes the infection potential of this dose. The necessity for the large dose in establishing infection can be explained either by the low probability for infection by a single organism, the hypothesis of independent action, or by the necessity for synergic action between individual infective organisms—the hypothesis of synergistic action.

Evidence available from plant pathology supports that from animal pathogens and accords better with the hypothesis of independent action. However, the possibility still exists that synergism may be involved in the production of disease in an 'unnatural' host, for example, where it is necessary to apply heavy doses of rust fungus spores to the leaf surface to produce disease. The chief evidence of synergistic action is provided by experiments with fungi infecting the roots of trees. The high resistance of the roots can only be overcome by a fungal inoculum provided with a food base, and infection is accomplished by mycelial strands and not by individual hyphæ.

Dr. G. Salt (King's College, Cambridge) spoke about the environment of larval parasitoid insects. The ecology of these parasitoids has two distinct aspects. The adults are free-living, usually winged, and the ways in which the females select the hosts and parasitize them have been widely studied. However, the ecology of the parasitoid egg or larva living on or in a particular host has received relatively little attention. Dr. Salt described some of his work on the life-history of a parasitoid larva within the hæmocœl of its larval insect host and gave an account of the environment, the changes which must occur in it if the parasite is to survive and the interactions between host and larva. An 'unnatural' host reacts to the presence of the larva in its hæmocœl, the larva becoming surrounded by an envelope of host blood which prevents development.

Dr. D. Arthur (King's College, London) described the ways in which ticks select their hosts and the feeding sites upon them. Hard ticks, on the tips of vegetation, are aroused to a state of preparedness by non-directional, tactile stimuli. These stimuli are reinforced by interacting odour and temperature stimuli prior to attachment to the host. In soft ticks, temperature appears to be the dominant stimulus for attachment.

The range of hosts selected by the parasite is not precisely known because of incorrect identification of ticks by earlier workers. However, the majority of ticks appear to be non-specific in their host selection. Some are limited to certain groups of hosts, but very few are specific to one host species, even among primitive ticks feeding on primitive mammals and the more advanced forms such as those found exclusively on Primates. For example, *Ornithodorus moubata* appears in three biological forms, that found on warthogs, porcupines and ant-bears possibly being primitive. The sites of attachment of various species and the reasons for their selection were discussed.

The chairman, Dr. E. L. Taylor (Veterinary Laboratory, Weybridge), opened the discussion. The adaptations of a parasite show various aspects operating simultaneously. The ecology of stages outside the host is similar to that for free-living organisms, but the parasite has also to find its host, effect an entrance or attachment and then behave in a way that ensures its own and its host's continued existence. The effect of parasite on host has been widely studied, but the effect of the host on the parasite should also be considered. Indeed, the two physiologies are interacting and in some circumstances mutual benefit may accrue. Contrary to the usual tenet of successful parasitism, the parasitoid insect larva kills its host, and Dr. Salt commented that the percentage of host population infected was small and losses, which could be considered as due to a refined form of predation, could easily be supported. Dr. Hindle cited examples of the extinction of host species by parasitism. Immunity reactions of the host, although prejudicial to an individual parasite, operate for the benefit of the species by limiting the parasite population. This mechanism is different in plant and animal hosts, and Dr. Jones said that plants do not show immune reactions, but rather a resistance to infestation which is a property of the individual cells. Parasite populations of plants show both density-dependent effects and independent factors, such as the steady rate of decay of the population of cyst-forming eelworms in the soil.

Immunity in invertebrates has been little studied, but there appears to be no true immunity although the production of non-specific antibodies may be evoked by the introduction of any foreign object. When the parasitoid insect larva with its envelope of host blood is transferred to another similar host there is no homograft reaction such as occurs in vertebrates.

Dr. Garrett suggested that another important difference between the plant and animal host is that in the plant there is always a region of young tissue at the growing points, whereas in animals growth usually ceases at maturity. Young animals are more susceptible than adults, and in plants the rootlets, for example, are more susceptible than the roots, the resistance of which can only be overcome by the most specialized of parasites. This phenomenon of age-resistance also operates in favour of the parasite by limiting the population.

The proceedings of this meeting will not be published in full. JUNE MAHON

ORIGIN OF TEKTITES

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CERTAIN aspects of recently advanced concepts relating to the origin of tektites require some considerable modification in the light of the conclusions arising from long-continued and detailed studies of the tektites from Australia. So far as is known, australites are the most recently arrived tektites on the Earth, and have evidently not lain upon the Earth's surface for more than 5,000 years. Furthermore, in terms of preservation of original shape types modified by secondary phenomena generated during rapid transit through the Earth's atmosphere, they are unmatched by any tektites from any other strewn-field on the Earth. Consequently, they are in the best condition on which to base conclusions as to their origin. Moreover, the very best preserved, most perfectly developed australites, which come from the Port Campbell district on the southern coast of Western Victoria, are retained in Australia, and have not been critically examined by overseas writers who make reference to australites in their publications.

Much detailed information has now accrued relating to the primary shapes, the secondarily modified shapes arising from the action of aerodynamical phenomena during ultra-supersonic and supersonic flight through the Earth's atmosphere, and the effects of tertiary phenomena such as subaerial erosion resulting from 5,000 years exposure to atmospheric agents. Considerable and detailed studies of the specific gravity of the same and different types of shape from the same and from different localities have been carried out. Many refractive index determinations have been made and specific refractivities calculated. Arcs and radii of curvature of remnant primary (posterior) and newly formed secondary (anterior) surfaces have been determined graphically from silhouette tracings, and significant conclusions drawn from these. Several etch solution tests have been conducted which provide valuable information of the effects of natural etching upon original structural elements and sculpture patterns of australites. Three more chemical analyses have been made and the composition related to specific gravity and refractive index values. The effects of heating to 1,200° C. has added further knowledge to the australite problem generally.

Studies of Darwin glass, 'straw-silica glass' and other non-volcanic terrestrial glasses have led to the rejection of Darwin glass from the group of the true tektites. Thin-section studies of the internal schlieren of australites have clarified the relationships between the core or body portion and the circumferential flange. All the structures, sculpture patterns and present shapes of australites can be adequately explained in terms of the few primary forms developed in an extraterrestrial environment, and subsequent modification by aerodynamical phenomena during high-speed earthward flight through the Earth's atmosphere, with a further modification by fragmentation, abrasion and solution-etching of the shapes so developed.

The results of these and other detailed studies have appeared during the past two decades[1].

The question of the origin of australites is admittedly still unsolved, but the weight of evidence from Australian studies is strongly in favour of an extraterrestrial source.

The concept has recently been advanced[2] that the arrival of tektites from space, with the distribution found on Earth, is not possible. Urey has calculated that if australites arrived as a swarm, its mean density must have been greater than 10^{-6} gm. cm.^{3}, its diameter some 10^{8} cm., and hence there would be a piling up of australites to a depth of 100 gm. cm.$^{-2}$ over southern Australia. Urey further states that he knows no answer to this problem. Assuming that the mean density and diameter of the swarm of australites are those given by Urey, detailed studies of australites and their distribution immediately supply the answer to the problem of their piling up to the depth suggested. As Urey states, this piling up is certainly "not the case". In fact, such a piling up to the depth calculated never could occur, for three very significant processes have operated to prevent it. The first and most important process operated for a very short period of time, but was, indeed, most effective in reducing the bulk of the swarm. The australites provide all the necessary evidence to show that they entered the Earth's atmosphere as small, cold (and hence solid) bodies with pre-formed primary shapes and pre-determined sculpture patterns, and that during high-speed transit through the atmosphere, in a matter of minutes at most, many of these forms were reduced in bulk by up to and more than 65 per cent, some by up to 80 per cent. This arose from the development of shock waves causing high pressures and temperatures, the generation of laminar boundary layer flow and reversed boundary layer flow leading to fusion stripping and ablation of the leading surfaces of the australites, and consequently considerable reduction in the size of each original form. Here, then, is an excellent means of reducing the estimated figure of a piling up process by two-thirds and more, and the amount lost in this way must be even greater when it is considered that an untold number of smaller original forms (less than 0·5 cm. in diameter) were undoubtedly completely consumed during atmospheric flight, as evidenced by studies of the smallest complete forms that survived the processes of ablation and thin-film fusion-stripping.

Urey has expressed surprise that a small glass object 1–2 cm. in diameter would travel at high velocity through the atmosphere, keeping one orientation as is necessary in the case of the more button-shaped tektites. Nevertheless, australites have done just this, and, moreover, not the "more button-shaped" examples alone, but also the ovals, boats, dumbbells, teardrops and canoes, etc., the better preserved examples of which all reveal identical types of secondarily developed structures and features (for example, circumferential flanges, concentric flow-ridges, clockwise spiral flow-ridges, anticlockwise spiral flow-ridges, radial flow-lines on the anterior surfaces, and so on). The role of aerodynamical phenomena at high speeds of flight supplies the answer to the development of all these features, and nearly all the australites bear testimony to the maintenance of a steady state of airflow, a state that can only be achieved during supersonic (and ultrasupersonic) speeds of atmospheric flight, with translatory forward motion only, and neither rotation about a polar axis, nor overturning during flight. As long as supersonic airflow obtains, a state of steady flight is maintained; but evidence is present on the anterior surfaces of some australites to indicate that although the motion of the objects was definitely limited to one direction, there arose some degree of 'wobbling' as an outcome of form-drag and buffeting at the high speeds that prevailed. Whether any of the australites in the swarm were affected by accidental or overtake collisions, it has not yet been possible to prove.

After australites landed on the Earth's surface, many without fracturing on impact, processes of abrasion and solution-etching played their part throughout the next 5,000 years or so, thus further reducing the mass of many forms. This is confirmed from detailed studies of the curvature – size relationships of excellently preserved australites, taken in conjunction with studies of other australites that reveal a whole range of tertiary modifications by subaerial agencies, from slightly worn and etched shapes that are almost perfect, right through to strongly abraded and/or deeply etched, now incomplete specimens, and all manner of fragments that can be assigned to one or another of the few shape groups, as well as many nondescript fragments. On the whole, australites have not lost anywhere near the amounts of glass by abrasion and etching as have tektites from other strewn-fields in other parts of the world, such as bediasites (which are very much older in terms of the time they have been on earth surface) and rizalites, etc. (which are also older, but also occur in regions subject to more pluvial, tropical conditions). Nevertheless, significant amounts of abrasion and solution-etching have been effective in certain parts of the Australian strewn-field, more especially in the arid regions and marginal lands, as is evident from comparisons between specimens from those regions and specimens in better preserved states from the more temperate regions of the island continent.

However, it is at present impracticable to assess the amount of australite glass lost from abraded specimens or the amount removed in solution by etching, for the whole of the 2 million square mile strewn-field. All that can be said with certainty at present is that the examination of some 20,000 specimens of australites in various states of preservation from various localities within the strewn-field indicates that between 50 per cent and 75 per cent of these have lost some 5–10 per cent of the original form, some by attrition from sand-blasting in certain areas, others from rolling in stream beds, some by solution-etching in certain soils, some by both processes, according to their location in the vast strewn-field. Several specimens of australites that are badly worn or deeply etched have lost 50 per cent or more of the original form. Furthermore, studies of the etching of smaller forms and small fragments lead to the conclusion that an unknown number has been completely destroyed by processes of subaerial erosion.

Current etching experiments with australites reveal that a specimen weighing approximately 2 gm. passes completely into solution in 4 per cent hydrogen fluoride in approximately 1,180 hr. at room temperatures. The rate of solubility was 3–4 mgm./hr. during the initial 300 hr. immersion, falling to less than 0·5 mgm./hr. after 1,000 hr. immersion, at which stage the initial form had been reduced in bulk by approximately 99 per cent. Although soil solutions are expected to be far less potent than 4 per cent hydrogen fluoride, nevertheless such solutions have been operative in varying concentrations and in varying quantities from place to place for some 5,000 years. In the most favourable circumstances, therefore, it is apparent that some australites in the right environment where solution-etching has been prevalent have been much reduced in bulk as a consequence of the removal of soluble ions. That entire forms of australites can have been completely dissolved by such a process is indicated by the 2-gm. weight australite that dissolved in 4 per cent hydrogen fluoride in 1,180 hr. without leaving any insoluble residue whatsoever.

Here, then, is a second important factor responsible for causing australite distribution to appear as it does to-day, and it is thus a significant factor in further reducing Urey's estimate of what the thickness of australites should be on the Earth's surface if entering from outer space as a shower; originally the thickness was somewhat greater than now, but loss by subaerial agents of erosion acting for some 5,000 years has been of considerable importance.

A third factor of possibly lesser importance than the first two, but nevertheless of some significance, affects the present distribution of australites on the surface, and hence, indirectly, the thickness of the australite 'pile up' from place to place. This factor introduces the operation of transporting agents such as streams, birds and aboriginal man. It has been generally accepted in the past that australites are normally found in the places where they originally fell to Earth. This seems to apply in many parts of the strewn-field, but in others there is indisputable evidence of movement by stream action into alluvial tin and alluvial gold deposits, as well as into clay pans and lake beds. I have observed several that have been washed over the edges of steep marine cliffs by surface run-off; these fall into talus cones and beach deposits and are ultimately lost by abrasion or swept out by tides on to the adjoining sea-floor. Others have been observed in stream beds entering the sea at grade, and thus provide a pointer to the fact that many specimens that become washed into water-courses ultimately are carried out on to the sea floor from farther inland.

It is a well-known fact that the Australian emu and the plain turkey utilize australites of suitable shape and dimensions as gizzard stones, and thereby can transport australites over considerable distances, at the same time subjecting them to marked degrees of wear in their function as 'grinder-stones'; these birds have been doing this sort of thing since australites first became available to them on the surface of the Earth, as evidenced by the discovery of gizzard stones with the fossilized remains of their ancestors. Aboriginal man has further added to the problem of present australite distribution, inasmuch as he prized these tektites as objects of magic and mystery, and carried them around on his nomadic journeyings; this is further evidenced by the discovery of australites in ancient aboriginal camps and kitchen middens, and by the fact that some show signs of 'carry polish' while others have been fashioned into crude, small implements.

Such events in the post-atmospheric flight history of australites must undoubtedly be taken into account when considering the initial scatter and thickness of australites in the strewn-field. There is no shadow of doubt that in many places the original distribution was by no means what it is to-day.

Thus, the three factors: (*a*) ablation and fusion stripping of primary forms during ultrasupersonic and supersonic earthward flight through the atmosphere; (*b*) subsequent abrasion, fragmentation and solution etching by subaerial agents; (*c*) the habits and activities of certain native birds and aboriginal man; have all been potent in affecting[1] the amount of australite glass that ultimately reached the Earth's surface, and then[2] in affecting its distribution and thickness on the surface. They all add up to a logical reason for the fact that australites are not now piled up to the calculated thickness of 100 gm. cm.$^{-2}$ if they came in from outer space as a shower, and with this objection to the present known thickness removed, added to the knowledge that australites show all the marks of having been subjected to the action of aerodynamical phenomena at high speeds of earthward flight downwards only through the atmosphere, there should be no really serious objection left to the entry of australites from outer space as a shower. Inasmuch as the other tektites from the other known seven strewn-fields of the Earth are much older, much more abraded or infinitely more affected by solution-etching, and do not reveal the remarkable features possessed by australites, it is at present impossible to extend *in toto* these ideas to explain their origin.

Further to the problem of tektite origin, Urey has set out reasons for regarding the several puzzling features of tektites as being capable of explanation in terms of derivation from cometary collision. In fact, however, the reasons advanced only serve to confuse the issue even more than it has been confused in the past several decades. The same arguments apply against the chances of cometary collision being the cause as arguments already advanced[1] against the much earlier theories such as Spencer's meteorite splash hypothesis[3] and Kahn's contraterrene meteorite impact theory[4].

Fatal to all three theories are the facts that the eight accepted types of true tektites fell to the Earth at widely separated times in late- to post-Mesozoic Earth history, and hence cannot possibly fit into the scheme of one cometary collision every 50 million years. The 50 million year cometary cycle was evidently designed to produce bediasites in the Eocene and australites, etc., in the Pleistocene.

The geological evidence, however, shows that the moldavites are Middle Miocene, the Ivory Coast tektites have been suggested to be late Mesozoic, while the bediasites were found in Pleistocene gravels and said to be derived from the Eocene. There are time-intervals of 10–30 million years between these assigned ages. Later in Earth history, the indomalaysianites are regarded as Middle Pliocene, the javaites as Middle Pleistocene, the rizalites as middle or early part of the Late (? Upper) Pleistocene, while the australites are undoubtedly the youngest of all, and definitely Holocene.

There are time-intervals of 3 or 4 million years between the groups of younger tektites, and time-intervals of 15–20 million years between them and the Miocene moldavites. It is thus apparent that the idea of tektite generation in 50 million year cycles of cometary collision cannot be accepted as it stands. Moreover, such cometary collisions would have to result in the meagre total of 2,000 known bediasites by the Eocene cometary collision, produced from a very restricted part of the Earth's surface, and at the other end of a 50-million year cycle, another cometary collision would be required to produce about 600,000 known australites, rizalites, javaites, billitonites and indomalaysianites spread over thousands of square miles of territories widely separated from one another.

Even abrasion, fragmentation and solution-etching differences between the results of the first cometary collision and the second could scarcely be expected to account for such a discrepancy, and at all events, the big Indomalaysian–Philippines–East Indies–Australian strewn-fields contain tektite types that fell to the Earth at different periods of late Tertiary to Quaternary Earth history. A single cometary collision in the Pleistocene cannot therefore explain the origin of the tektites ranging in age from Middle Pliocene to Holocene in the Far East–Australian region. Such a high-energy explosion as that postulated to have affected so vast an area of the Earth's surface must surely have had greater and more far-reaching effects than the mere production of a few thousand pieces of glass of relatively small size; no such effects have been recognized or postulated.

A further difficulty with such a suggested origin is that the single high-energy explosion would be required to fuse all manner of terrestrial materials of different composition, so as to produce tektite glass of generally similar chemical composition in areas 65° of latitude apart. Even if the cometary collision supplied the energy for fusion, the wide array of superficial terrestrial materials would by no means supply relatively homogeneous tektite glass, even with the postulated fractionation into acid and basic fractions, a process which, in itself, is most difficult to conceive during the upward propulsion of fused Earth material that afterwards re-entered Earth's atmosphere according to the cometary collision theory. There should, moreover, be a whole range of partially fused materials formed at each milieu affected by the cometary collision; there should also be a whole range of different kinds of glass irrespective of the postulated acid and basic glasses; there should be removal of the basic glasses (or else they have never been found), leaving only the acid tektite glass. Furthermore, this one postulated cometary collision would have to be responsible for shaping the different types of tektites in different ways to produce different shape types at widely separated strewn-fields, and would, in some unexplained manner, have to return them to Earth at intervals separated by some 3 or 4 million years. Another cometary collision would then be necessary to explain the Middle Miocene moldavites, and so on, with the result that as many cometary collisions would need to be postulated, and all within the limits of the originally suggested 50 million year cycle of cometary collision, as there has been suggested Great Circles to account for tektite distribution. In other words—a meaningless maze.

Finally, the australites are not consonant with the suggestion of fusion of Earth substance by a high-energy explosion, propulsion to great heights (beyond the atmospheric envelope) with the production of primary forms, and then return to Earth through the atmosphere to generate the secondary shapes.

Admittedly high-speed transit through the Earth's atmosphere has produced the secondary shapes from primary forms. It is not agreed, however, that the composition of australites can be explained in terms of the fusion of the kind of materials exposed at the Earth's surface, and at the same time show the type of chemical gradient according to a provincial distribution across 2,000 miles of the island continent. There is no evidence whatsoever that australites received their primary shapes by upward propulsion from the Earth's surface. None of the known shape types reveal the characteristics shown by volcanic bombs, for example, which are propelled in a plastic state, sometimes to considerable heights, although never, as far as known, high enough to leave the Earth's atmospheric envelope. All the evidence so far accrued is convincing that the australites received their secondary shapes by the modification of primary spheres, spheroids, dumbbells of revolution and apioids of revolution that entered the Earth's atmosphere as solid, non-rotating bodies at ultra-supersonic velocities, traversed the atmosphere at lesser speeds nearer the Earth's surface, passing through a supersonic zone, through the transonic zone and finally into the subsonic zone. During this transit, their leading surfaces were sculptured into the shapes and designs now known from studies of the very excellently preserved, perfectly developed specimens from the Port Campbell district of Western Victoria.

All the evidence points to the fact that the majority of these forms maintained stable positions during high-speed flight through the atmosphere, and while supersonic or higher speeds prevailed, they always presented one surface only to the effects of the aerodynamical phenomena responsible for their secondary sculpture. Moreover, each entity in the swarm, whether original sphere, spheroid, dumbbell or apioid, or even hollow form, behaved in unison with the rest, and each received identical treatment from the medium through which it had its trajectory, so that identical secondary features were produced on the forward surface of each shape type, such features only varying in minor degrees according to the shape types on which they were produced. No secondary shapes have been found that would indicate modification of such geometrically possible forms of revolution as the annular torus and the paraboloid; but this does not mean that they were not present. Some of the so-called 'aberrant' types of australite shapes, which have been subjected to the secondary modification by aerodynamical phenomena in the same way as the better known shape types, were undoubtedly pre-formed shapes before entry into the Earth's atmosphere from outer space. They may have been originally of the paraboloid or annular torus shape types that suffered accidental collapse or other modification in their extra-terrestrial birthplace.

In the face of all these problems and difficulties that beset research work on tektites, I cannot but conclude that cometary collisions spaced at 50-million year intervals are entirely incapable of satisfactorily explaining tektite origin, or of solving the present distribution of widely spaced strewn-fields containing different shape types of tektites that fell to Earth at widely separated intervals. The position still stands as it did approximately seventy years ago, when it was stated about tektites (then referred to as "obsidianites")—"where they came from no one knows". Research since then has shown that for the australites at least, an extra-terrestrial origin of tektite glass is by far the most likely, but it is still unknown whence in extra-terrestrial space the tektites were derived.

[1] Baker, G., *Proc. Roy. Soc. Vic.*, **49** (2), 165 (1937); *Min. Mag.*, **25**, 487 (1940); *Proc. Roy. Soc. Vic.*, **52** (2), 312 (1940); *Mem. Nat. Mus. Vict.*, **14** (1), 7 (1944); **14** (2), 47 (1946); *Proc. Roy. Soc. Vic.*, **60**, 35 (1950); *Trans. Roy. Soc. Sth. Aust.*, **76**, 1, 27 (1953); *Proc. Roy. Soc. Vic.*, **67**, 165 (1955); *Min. Mag.*, **30**, 596 (1955); *Mem. Nat. Mus. Vict.*, **20**, 59, 173 (1956); **21**, 72 (1957); **22**, 1 (1957); *Amer. J. Sci.*, **256**, 369 (1958); *Mem. Nat. Mus. Vict.*, **23**, 5 (1959); **24**, 69 (1959). Baker, G., and Forster, H. C., *Amer. J. Sci.*, **241**, 377 (1943). Baker, G., and Gaskin, A. J., *J. Geol.*, **54**, 88 (1946).

[2] Urey, H. C., *Proc. U.S. Nat. Acad. Sci.*, **41**, 27 (1955); *Nature*, **179**, 556 (1957).

[3] Spencer, L. J., *Nature*, **131**, 117 (1933).

[4] Kahn, M. A. R., *Contrib. Met. Soc.*, **4** (1), 35 (1947).

[See also p. 300 of this issue.—EDITORS.]

LIPOAMINO-ACIDS: THE QUESTION OF ARTEFACT

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THE name 'lipoamino-acid' is proposed for a lipid molecule as normally defined[1] from which one or more amino-acid molecules can be cleaved. For lipoamino-acids in which peptide bonding is known to occur the name 'lipopeptide'[2-7] is appropriate.

There are ever more numerous reports of the existence of lipoamino-acids in lipid samples prepared from many biological materials. Samples demonstrably uncontaminated with free amino-acids or peptides afford various amino-acids on acid hydrolysis. The amino-acids appear to be in covalent linkage, since lipoamino-acids have survived at a wide range of pH values (without heat) and during treatment with salts under various conditions, solvent fractionation (including countercurrent distribution), electrodialysis, electrophoresis, paper chromatography, and column chromatography on alumina, cellulose, 'Hyflo Supercel', and silicic acid. They have even survived treatment with proteolytic enzymes, Van Slyke deamination[8], and dialysis with *N* hydrochloric acid for three days[9].

One lipoamino-acid, phosphatidylserine, has been isolated pure, and its chemical structure is established. Others have only been revealed as minor constituents of mixtures, which in a few cases contained no phosphorus. Their structures cannot be analogous to that of phosphatidylserine, particularly because in many samples hydroxy amino-acids were absent or were present only in small proportions.

Some samples containing lipoamino-acids were devoid of free amino-groups, as indicated by negative ninhydrin tests. For example, in my hands lipid fractions from blood, egg, flies, liver, and milk, eluted from silicic acid by 5 per cent (v/v) methanol in chloroform, under no conditions gave more than a faint yellow ninhydrin colour on heavily loaded paper chromatograms. The sodium 1,2-naphthoquinone-4-sulphonate test[10] was also negative, yet hydrolysis liberated amino-acids. Total amino-acids (chiefly glutamic) liberated from the fly (*Drosophila melanogaster*) fraction[11] amounted to 0·07 mole per kilogram of lipid. They were liberated at 105–110° by alkali as well as by acid, but not by water, and not at atmospheric temperature and pressure by hydrogen with Adams catalyst.

Peptide bonding could account for absence of free amino-groups, but was ruled out for the above fractions, and for some others, by negative chlorination tests[11] and by absence of 'Amide II' absorption in the infra-red (unpublished experiments).

Detection of amino-acids after hydrolysis of lipid preparations does not *necessarily* indicate the occurrence of peptide or protein moieties, and claims in the literature that it does should be viewed critically.

During examination of lipids from several sources by silicic acid chromatography it was noticed that commercial dried blood and egg, and decaying *Drosophila*, gave fractions with higher and more varied amino-acid content than sources believed free from non-enzymic and/or enzymic deterioration. It therefore seemed possible that lipoamino-acids might be artefacts not existing *in vivo*. Binding of nitrogenous substances is a well-known if ill-understood concomitant of lipid autoxidation[12,13], which might occur during extraction and manipulation.

Experiments with linseed oil supported this hypothesis. Raw linseed oil was freed of phospholipids and other contaminants by passage in chloroform through a silicic acid column. (It still contained minute traces of lipoamino-acids.) This oil (10 ml.) was mixed with water (10 ml.), glycine (0·25 gm.), and hæmoglobin (0·1 gm.) in a large test-tube into which a brisk stream of filtered air was passed for 2 days at 25°. The chloroform-soluble portion of the product was chromatographed on silicic acid. Material eluted by pure chloroform afforded only traces of amino-acids on hydrolysis, but material (ninhydrin-negative) eluted by 10 per cent methanol in chloroform afforded approximately 2 mgm. of glycine. Purification by dialysis, instead of by chromatography, confirmed that silicic acid was not responsible for this result. Unreacted, contaminating glycine was only eluted from the column by pure methanol[11]. Leucine and serine, used in place of glycine, gave similar results. When hæmoglobin was omitted from the reaction mixtures, or when the oil was subjected to normal conditions of lipid extraction, no binding of amino-acids could be detected.

However, being less prone to autoxidation than highly unsaturated phospholipids, linseed oil is perhaps not a good model substance.

The following experiment was therefore made. A solution of glutamic acid uniformly labelled with carbon-14 (156 000 counts/min., weight negligible) in moist *n*-propanol was mixed with powder, prepared from *Drosophila* (15 gm.) and dry ice[11], before propanol extraction. The crude extract contained 105,000 counts, 41,000 being retained in ligroin-soluble material (0·82 gm.) which was then chromatographed on silicic acid (12·5 gm.). Specific activities of eluted fractions (in which > 90 per cent of counts were recovered) were determined using thin samples on aluminium planchettes. Results are shown in Table 1. That radioactivity was bound in lipid was demonstrated by paper electrophoresis[3]. Material eluted by 15 per cent methanol in chloroform was placed on a heavily loaded zone of non-radioactive glutamic and 2-pyrrolidone-5-carboxylic acids before electrophoresis. Afterwards, at least 85 per cent of the counts were found in the lipid zone at the origin. Confirmation was provided by 'three-run' paper chromatography (Fig. 1). When lipid eluted by 15 per cent methanol was thus tested zone *U* contained 22 per cent of the counts, *V* + *W* contained 62 per cent, and *X* + *Y* contained 16 per cent. Percentages for lipids eluted by 25 per cent methanol and by 35 per cent methanol were respectively 21, 67, 12 and 27, 30, 43. Zones *U* to *Y* contained fluorescent zones of autoxidation products, which probably accounted for the considerable losses of radioactivity from lipid zones. Significantly, lipid zones did retain more than 20 per cent of their original counts.

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Results also shown in Table 1 are of an experiment in which a considerable weight (1·0 gm. for 8 gm. *Drosophila*) of arginine hydrochloride (372,000 counts/ min.) was used instead of the negligible weight of glutamic acid.

Clearly, some lipoamino-acids are artefacts formed *in vitro* from endogenous amino-acids ; indeed, apart from phosphatidylserine, all of them might be artefacts. This possibility was tested by a series of experiments in which large amounts of selected amino-acids and other compounds ('additives') were added, singly or in pairs, to render concentrations of endogenous compounds insignificant from the beginning of extraction. Extraction and removal of non-lipid contaminants were carried out at low temperature[11].

In a typical experiment blood (50 ml.), collected from a horse, was within 2 min. poured into methanol (100 ml.), plus powdered dry ice (excess), plus additive (10–40 gm. per gm. total lipid). In a room at – 15° lipids were then extracted (total solvent amounted to 400 ml. chloroform–methanol, 2 : 1, v/v) and washed four times by the Folch (1957) sodium chloride procedure[11]. After evaporation of solvent (< 40°) the whole lipid (0·17–0·20 gm., all ligoin-soluble) was chromatographed on a silicic acid column (7·5 gm.). Materials eluted successively by chloroform (60 ml.), by 5 per cent (60 ml.), 15 per cent (240 ml.), and 25 per cent methanol in chloroform (150 ml.), and by methanol (80 ml.) were hydrolysed, and the hydrolysates were examined by paper chromatography[11]. Cow's milk, within 3 min. of milking, was handled similarly, except that additive in aqueous solution was mixed with it immediately before pouring into methanol. Whole *Drosophila* were powdered with dry ice plus additive, as mentioned above. Additives employed were: 2-amino-octanoic acid ; arginine hydrochloride ; ethyl vinyl ether ; guanidine hydrochloride ; hydroxyproline ; mercuric acetate ; monosodium glutamate ; potassium chloride ; sequestric acid ; serine ; α-tocopherol ; urea ; 2-amino-octanoic acid plus ascorbic acid, ethyl vinyl ether, hydroquinone, sequestric acid, tetrahydrofuran ; decylamine plus acetic acid ; glycine plus acetic acid, sequestric acid ; serine plus ethyl vinyl ether, guanidine hydrochloride.

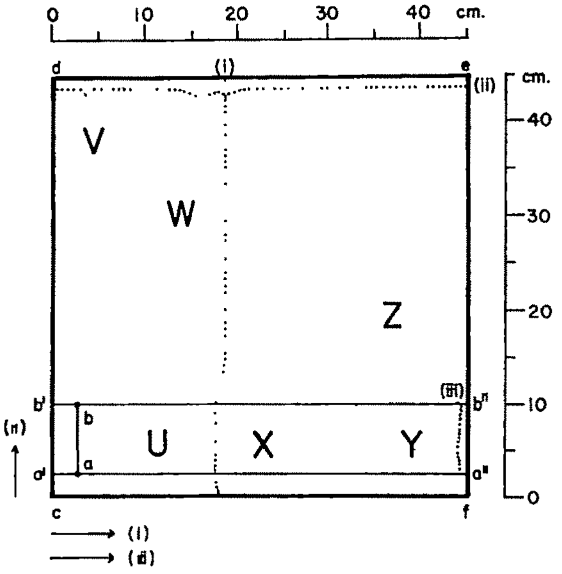

Fig. 1. 'Three-run' paper chromatography, a test for radioactive glutamic acid bound to lipid. Lipid was deposited on a line, *ab*, on a sheet of Whatman No. 3*MM* paper, *cdef*, and chromatographed (i) in benzene/*n*-butanol/methanol/water (1 : 1 : 2 : 1, v/v). After development in the second dimension (ii) with water, the strip *a'a''b'b''* was cut out, and heavy loads of non-radioactive glutamic and 2-pyrrolidone-5-carboxylic acids were applied to *ab* before development (iii) with water. Development was ascending. Numbered, dotted lines represent respective solvent fronts. Lipid formed a zone at *U* and the two acids at *Y*. The paper was treated with ninhydrin, and ultimately cut into several pieces, designated *U–Z*. These were eluted with 25 per cent water in *n*-propanol for radioactivity measurements

Table 1. INCORPORATION OF RADIOACTIVE AMINO-ACIDS INTO *Drosophila* LIPIDS DURING EXTRACTION AND MANIPULATION

Eluant	Counts/mgm./min. of eluted material	
	Glutamic acid	Arginine
$CHCl_3$	0	0
5 per cent MeOH	9 46	1
15 ,, MeOH	7	1 7
25 ,, MeOH	7	7
35 ,, MeOH	116	7
50 ,, MeOH	83	9
MeOH	1,510*	42*

* High values due to elution of the free amino-acids[11].

In control experiments hydrolysates of all but pure chloroform eluates showed fairly constant patterns of amino-acids (for typical patterns see refs. 11 and 14). In many of the additive experiments the patterns were little changed, and in the rest they were changed but not absent. Artefacts formed by several amino-acids, and by decylamine, were detected in amounts comparable with amounts of the usual lipoamino-acids present. For example, in experiments with 2-amino-octanoic acid, this acid appeared in hydrolysates of all except pure chloroform eluates ; it became bound in *Drosophila* lipids if added at any stage before chromatography.

Antioxidant additives failed to prevent the appearance of lipoamino-acids or the formation of artefacts. 2-Amino-octanoic acid formed large amounts of artefact in presence of ethyl vinyl ether, suggesting that vinyl ether groups of the naturally occurring plasmalogens may be involved in artefact formation. Lipoamino-acids do not themselves appear to be plasmalogens, since treatment of whole horse blood lipid with 90 per cent acetic acid at 65° before chromatography did not significantly alter the amino-acid pattern in hydrolysates.

Neglecting the unlikely possibility that in the experiments described above additives failed to equilibrate with endogenous amino-acids, it is concluded that, while some lipoamino-acids are artefacts, others do exist *in vivo*.

Grateful acknowledgement is made to Prof. H. K. Mitchell and Sir Alexander Todd for encouragement, to the Directors of J. Lyons and Co., Ltd., for facilities, and to the Rockefeller and National Science Foundations for financial support.

[1] LeBaron, F. N , and Folch, J., *Physiol. Rev.*, **37**, 539 (1957).

[2] Baer, E., "Ann Rev. Biochem.", **24**, 135 (1955).

[3] Westley, J., Wren, J. J., and Mitchell, H. K., *J. Biol. Chem.*, **229**, 131 (1957).

[4] Grady, J. E., Koffler, H., Parsons, J. L., Kobayashi, T., and Tetrault, P. A., *Fed. Proc* , **17**, 233 (1958).

[5] Guinand, M., Michel, G., and Lederer, E., *C.R. Acad. Sci., Paris*, **246**, 848 (1958)

[6] Wallach, D. F. H., and Garvin, J. E., *J. Amer Chem. Soc* , **80**, 2157 (1958).

[7] Klenk, E , and Debuch, H., "Ann. Rev. Biochem.", **28**, 39 (1959).

[8] Hecht, E., and Mink, O., *Biochim. Biophys. Acta*, **8**, 641 (1952).

[9] Hawthorne, J. N., and Chargaff, E., *J. Biol. Chem.*, **206**, 27 (1954).

[10] Giri, K. V., and Nagabhushanam, A., *Naturwiss* , **39**, 548 (1952).

[11] Wren, J J , and Mitchell, H. K , *J. Biol. Chem* , **234**, 2823 (1959).

[12] Narayan, K. A., and Kummerow, F. A., *J. Amer. Oil Chem. Soc.*, **35**, 52 (1958).

[13] Venolia, A. W., and Tappel, A. L., *J. Amer. Oil Chem. Soc.*, **35**, 135 (1958).

[14] Wren, J. J., and Mitchell, H. K., *Proc. Soc. Exp. Biol. Med.*, **99**, 431 (1958).

IS THERE A LEAKAGE OF ACETYLCHOLINE FROM POSTGANGLIONIC PARASYMPATHETIC NERVE ENDINGS?

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WHEN the chorda tympani is cut, a supersensitivity to chemical agents develops in the submaxillary gland, as first shown by Maevsky[1] and Fleming and MacIntosh[2]. A supersensitivity of similar type can be created by treating an animal for some weeks with ganglionic blocking compounds[3] or atropine[4]; since the sensitization is unspecific, secretory agents such as adrenaline can be used to show the effect of atropinization. These observations suggest that the gland cells acquire an increased sensitivity toward chemical stimuli when deprived of secretory impulses[5], and this hypothesis is supported by the finding that some supersensitivity can be produced by cutting afferent fibres of the reflex arc, the lingual or glossopharyngeal nerves[6]. As further evidence in this direction the fact may be quoted that the supersensitivity obtained after section of the chorda can be abolished by daily subcutaneous injections of secretory drugs such as pilocarpine or carbachol; even the responsiveness of the normally innervated gland can be lowered by this procedure[7].

A closer study of the effect of treatment with parasympatholytic agents reveals, however, that the transmitter, released by the nerve impulse, cannot be the only factor which determines the degree of sensitivity of the gland cells.

When atropine treatment is used to create supersensitivity, the doses of the drug have to be increased progressively since tolerance develops; if the initial small dose is given for a long period the sensitivity will first rise, and then decline again in spite of continued treatment (Emmelin, N., and MacIntosh, F. C., unpublished work, 1955). It is therefore difficult to compare the maximal level obtainable by atropinization with that brought about by section of the chorda. By using another parasympatholytic agent, piperidino-ethyl-diphenyl-acetamide (Hoechst 9980), it is possible to produce a marked supersensitivity without increasing the dose of the drug[8]. The supersensitivity obtained in such experiments is found to be much higher than that following section of the chorda[9]. This drug is very specific as a parasympatholytic agent; in the doses used it has, for example, no ganglionic blocking action and does not change the sensitivity to adrenaline in the acute experiment. It therefore seems reasonable to assume that it exerts its sensitizing action by virtue of its parasympatholytic properties. The supersensitivity caused by this drug should then in part be due to the fact that this agent, just as section of the chorda, deprives the gland of the action of acetylcholine liberated by secretory impulses from the central nervous system; but the sensitization above the chorda-level suggests that the drug in addition abolishes some action of a cholinergic mechanism which is left intact when the chorda is cut. In order to test the hypothesis that the supersensitivity caused by Hoechst 9980 is entirely due to loss of action of acetylcholine on the gland cells, some other parasympatholytic agents have been tried: methylscopolamine, lachesine and isopropamide (diphenylcarbamoyl - propyl - di*iso*propyl - methylammonium iodide). They all increase the sensitivity of the gland, all to the same level and to the level characteristic of treatment with Hoechst 9980 (unpublished work).

Section of the chorda tympani is a preganglionic denervation. It leaves the parasympathetic, cholinergic, postganglionic neurone intact. The obvious experiment would be to cut these fibres and investigate whether a supersensitivity ensues which is of the same size as that produced by treatment with parasympatholytic agents. Unfortunately, some of the synapses of the parasympathetic fibres are situated within the gland and not anatomically accessible. Many, however, are found close to the gland but still outside it, and by dissecting the nerve towards the hilum it is possible to produce a partial postganglionic parasympathetic denervation. A supersensitivity is found to develop which is not as high as that produced by treatment with parasympatholytic drugs but more pronounced than that following section of the chorda (unpublished work). Care has to be taken not to damage the postganglionic sympathetic fibres; this in itself would cause a supersensitivity which can be superimposed upon that due to section of the chorda[10]. In the parotid gland of the cat, it is feasible to cut all the postganglionic parasympathetic fibres by section of the auriculotemporal nerves. This operation produces a supersensitivity towards acetylcholine which is larger than that caused by preganglionic denervation[11]. It is, however, not possible to make comparisons between this sensitization and that produced by treatment with parasympatholytic agents because of lack of a suitable test drug; acetylcholine cannot be used since its secretory effect is abolished by such drugs. Adrenaline has to be given in large doses to produce a secretion from the parotid gland, and interfering vascular effects render quantitative comparisons impossible.

The question arises whether there is any indication of an activity in the postganglionic, parasympathetic neurone, apart from that due to impulses from the superior salivary nucleus, which results in a release of acetylcholine. There are two observations which could be quoted in support of the view that small doses of acetylcholine are continuously liberated from the endings of the postganglionic neurone, even when the chorda has been cut.

First, when the submaxillary gland is perfused with an eserinized solution, very small amounts of acetylcholine can be detected in samples of perfusate collected when the chorda has been cut acutely but is not stimulated[12]; this is the case even when the perfusion fluid contains curarine[13].

Secondly, eserine is known to cause a flow of saliva. This effect can be obtained when a small dose of eserine is injected intra-arterially near the gland[14] or through the duct towards the gland[15]. A very small dose, given through the duct, has no effect of its own but increases the secretory effect of injected acetylcholine. If the dose of eserine is raised, a flow of saliva is evoked even when no acetylcholine is administered. It is reasonable to connect this secretory action of eserine with an anticholinesterase effect. This is further supported by the fact that other cholinesterase inhibitors, like *TEPP* and paraoxon, cause a similar secretion. The secretory effect of these drugs is obtained after section of the chorda, and it is then as big as when the chorda is intact. If the secretion is due to acetylcholine, preserved by the esterase inhibitors, this acetylcholine has therefore not been released by impulses from the central nervous system. The secretory effect of eserine remains when the chorda fibres have been cut in advance and allowed to degenerate; after about a week or more it is even larger than for the normally innervated gland because of the sensitization of the gland cells.

Corresponding results are obtained in the parotid gland; but in this preparation it is possible to analyse further the secretory action of anticholinesterases[16]. When the postganglionic parasympathetic fibres have been cut and left to degenerate, eserine injected through the duct has only a very small secretory effect, in spite of the fact that the gland is even more sensitive to acetylcholine than after preganglionic section. Therefore, it seems legitimate to conclude that the main secretory action of eserine is connected with the appearance of acetylcholine originating from the endings of the postganglionic neurone. Such a release cannot be the result of impulses from the cell-body of this neurone, for the secretion is obtained when the postganglionic fibres have been severed acutely; nor is it likely, in such experiments, to be derived from impulses set up from the injured nerve stump, for the secretion is of the same size as that obtained from the contralateral normally innervated gland. A plausible explanation seems to be that there is a leakage of acetylcholine from the postganglionic parasympathetic endings similar to that which takes place at the somatomotor nerve terminals. It was, in fact, suggested already in 1945 by Feldberg[17] that there might be a more or less continuous release of small amounts of acetylcholine from the endings of cholinergic nerves in general. The amounts of acetylcholine released in this way should then be too small to cause any secretion; this would be the case even if the gland cells were sensitized by previous preganglionic denervation. On the other hand, if this acetylcholine should have anything to do with supersensitivity, it must in addition be assumed to exert some kind of action on the gland cells, the removal of which by postganglionic degenerative section or treatment with parasympatholytic agents manifests itself in supersensitivity. A higher level of supersensitivity after post- than after pre-ganglionic denervation is implied in Cannon's 'law of denervation'; leakage of transmitter at the postganglionic endings could provide an explanation for this phenomenon.

In the presence of an anticholinesterase the leaking acetylcholine must be assumed to accumulate to a concentration high enough to cause a flow of saliva. Such a concentration may be reached, under special conditions, even when no esterase inhibitor has been administered. One to three days after section of the postganglionic parasympathetic fibres of the parotid gland, a curious secretory activity can be found[18,19]. The flow is intermittent, occurring in bursts, and has been termed 'paroxysmal secretion'. Since it is abolished by atropine, it seems to be due to acetylcholine. It is, likewise, abolished by cocaine, injected through the salivary duct in a dose which does not change the responsiveness of the gland to injected acetylcholine; it is therefore likely that the acetylcholine responsible for the secretion is of nervous origin. It is reasonable to conclude that the acetylcholine is liberated from the endings of the degenerating nerve fibres. Since the flow persists when a local anæsthetic has been applied to the cut end of the nerve, the secretion cannot be initiated from that site. Acetylcholine seems to be released from the degenerating terminals in bursts in which the threshold concentration for secretion is temporarily surpassed.

[1] Maevsky, W. E., quoted by Babkin in "Secretory Mechanism of the Digestive Glands" (Hoeber, New York, 1944).

[2] Fleming, A. J., and MacIntosh, F. C., *Quart. J. Exp. Physiol.*, 25, 207 (1935).

[3] Emmelin, N., *Brit. J. Pharmacol.*, 14, 229 (1959).

[4] Emmelin, N., and Muren, A., *Nature*, 166, 610 (1950).

[5] Emmelin, N., *Physiol. Rev.*, 32, 21 (1952).

[6] Emmelin, N., abstr. Twentieth Int. Physiol. Cong., Brussels, 260 (1956).

[7] Emmelin, N., and Muren, A., *Acta Physiol. Scand.*, 26, 221 (1952).

[8] Emmelin, N., and Henriksson, K. G., *Acta Physiol. Scand.*, 30, Supp. 111, 75 (1953).

[9] Emmelin, N., and Stromblad, B. C. R., *Acta Physiol. Scand.*, 38, 319 (1957).

[10] Emmelin, N., and Muren, A., *Acta Physiol. Scand.*, 24, 103 (1951).

[11] Stromblad, R., *Acta Physiol. Scand.*, 33, 83 (1955).

[12] Hendersson, V. E., and Roepke, M. H., *Arch. Exp. Path. Pharmakol.*, 172, 314 (1933).

[13] Emmelin, N., and Muren, A., *Acta Physiol. Scand.*, 20, 13 (1950).

[14] Dirnhuber, P., and Evans, C. Lovatt, *Brit. J. Pharmacol.*, 9, 441 (1954).

[15] Emmelin, N., Muren, A., and Strömblad, R., *Acta Physiol. Scand.*, 32, 325 (1954).

[16] Emmelin, N., and Stromblad, B. C. R., *Brit. J. Pharmacol.* 13, 193 (1958).

[17] Feldberg, W., *J. Physiol.*, 103, 367 (1945).

[18] Emmelin, N., and Stromblad, B. C. R., *J. Physiol.*, 140, 21*P* (1958).

[19] Emmelin, N., and Stromblad, B. C. R., *J. Physiol.*, 143, 506 (1958).

LETTERS TO THE EDITORS

RADIOPHYSICS

Global Hydromagnetic Wave Ducts in the Exosphere

In 1957, one of us (H.A.B.) considered the initiation of an earth-magnetic field experiment to detect hydromagnetic waves in the upper ionosphere and exosphere. He suggested the use of very large wire loops (of about 100 km.^{2} area) as pick-up systems for earth-magnetic field perturbations. A large loop has the advantage of a very high signal-to-noise ratio, since it effectively averages out local magnetic interference.

Acting on this suggestion, Berthold and Harris of this Laboratory installed two wire-loop stations (in Arizona and New Jersey), which were operated simultaneously during the summer of 1958. In addition to the advantages due to the high sensitivity of the loops, very accurate synchronization of the two stations was possible. This accuracy made it possible for Berthold and Harris to measure, for the first time, exact time differences in the arrival of sudden commencements at different parts of the world[1].

During the *Argus* experiments of 1958, Berthold and Harris recorded, at both stations, magnetic signals that originated from the high-altitude detonations. Both stations recorded two signals: a fast one, which arrived with a travel time of about four seconds; and a slower one, which arrived about four to five times later than the fast signal[2]. Similar signals were also observed by an earth-current station operated by the U.S. Army Signal Research and Development Laboratory in Maine under the supervision of G. Carp. In addition, the same two signals were recorded during the *Argus* experiments by two stations (Uppsala, Sweden, and Reykjavik, Iceland) operated under the supervision of H. Benioff of the California Institute of Technology, and by a geomagnetic station operated by the U.S. Air Force at the Azores[3]. Table 1, column 2, gives the observed times of arrival of the slow signal at these six stations, and Table 2, column 2, gives the same results for the faster signal.

Table 1. Slow Mode (Transverse Hydromagnetic Mode)

	T (Experimental) sec.	$K \cdot \int f(\varphi)$	P	T (Theoretical) sec.
Arizona ..	26·0	$16{\cdot}55 \times 10^{-26}$	$1{\cdot}57 \times 10^{26}$	24·8
Uppsala ..	15·7	$11{\cdot}55 \times 10^{-26}$	$1{\cdot}36 \times 10^{26}$	17·4
Reykjavik ..	17·8	$12{\cdot}45 \times 10^{-26}$	$1{\cdot}43 \times 10^{26}$	18·7
Maine	17·7	$11{\cdot}10 \times 10^{-26}$	$1{\cdot}59 \times 10^{26}$	16·7
New Jersey .	18·3	$10{\cdot}90 \times 10^{-26}$	$1{\cdot}68 \times 10^{26}$	16·4
Azores .	10·8	$8{\cdot}00 \times 10^{-26}$	$1{\cdot}35 \times 10^{26}$	12·0

$K = \sqrt{4\pi}/H_0a^3 = 4{\cdot}58 \times 10^{-26}$ [gauss^{-1}.cm.$^{-3}$]
$\overline{P} = 1{\cdot}50 \times 10^{26}$
$P = R^4\sqrt{\rho}$ [cm.^{4} × (gm./cm.^{3})$^{1/2}$]

Table 2. Fast Mode (Longitudinal Hydromagnetic Mode)

	T (Experimental) sec.	$K \cdot \int f(\varphi)$	P	T (Theoretical) sec.
Arizona .	5·0	$8{\cdot}11 \times 10^{-26}$	$6{\cdot}17 \times 10^{25}$	5·3
Uppsala ..	4·3	$7{\cdot}10 \times 10^{-26}$	$6{\cdot}06 \times 10^{25}$	4·6
Reykjavik ..	4·6	$7{\cdot}05 \times 10^{-26}$	$6{\cdot}53 \times 10^{25}$	4·6
Maine ..	4·5	$6{\cdot}96 \times 10^{-26}$	$6{\cdot}48 \times 10^{25}$	4·5
New Jersey ..	4·7	$6{\cdot}92 \times 10^{-26}$	$6{\cdot}80 \times 10^{25}$	4·5
Azores .	4·2	$5{\cdot}95 \times 10^{-26}$	$7{\cdot}00 \times 10^{25}$	3·9

$K = \sqrt{4\pi}/H_0a^3 = 4{\cdot}58 \times 10^{-26}$ [gauss^{-1} . cm.$^{-3}$]
$\overline{P} = 6{\cdot}52 \times 10^{25}$
$P = R^4\sqrt{\rho}$ [cm.^{4} × (gm./cm.^{3})$^{1/2}$]

Arguing that the observed signals were caused by the propagation of hydromagnetic waves due to the sudden displacement of earth-magnetic force lines by the highly diamagnetic fireball, we arrived at the following conclusions: (1) the hydromagnetic waves at high altitudes are essentially confined in ducts, which in first approximation are concentric shells about the earth; (2) within these ducts, the hydromagnetic waves travel essentially along great circle paths; (3) the propagation modes in these ducts are the well-known transverse and longitudinal hydromagnetic-wave modes.

If ds is a line element of a great circle path in the assumed ducts, the travel time between two points is given by:

$$T = \int ds/v$$

If we tentatively identify the slow signal with a generalized Alfvén wave, then $v = v_a\cos\theta$, where $v_a = H/\sqrt{(4\pi\rho)}$, and θ is the angle between the field lines and the direction of propagation. The fast signal should then be identified with the longitudinal hydromagnetic wave, for which $v = v_a$, since in the upper atmosphere v_a is about one thousand times as great as the speed of sound.

Thus, we obtain the following expression for the generalized Alfvén mode of propagation through the duct:

$$T = R^4\sqrt{\rho}\cdot\frac{\sqrt{4\pi}}{H_0a^3}\cdot\frac{1}{2\sin\varphi_0}\left[\ln\frac{\sin\varphi_0+\sin\varphi_2}{\sin\varphi_0-\sin\varphi_2}-\ln\frac{\sin\varphi_0+\sin\varphi_1}{\sin\varphi_0-\sin\varphi_1}\right]$$

where R is the distance of the duct from the Earth (centre to centre); ρ is the ion mass density in the duct; $H_0 = 0{\cdot}3$ gauss (surface magnetic field-strength of the Earth at the equator); $a = 6{,}371$ km. (radius of the Earth); φ_1 and φ_2 are the magnetic latitudes of the site of detonation and receiver stations, respectively; φ_0 is defined as the maximum magnetic latitude of the great circle path, that is, the most northerly point of such a great circle. This value can be readily obtained by elementary trigonometrical computation from the values of φ_1, λ_1, φ_2, and λ_2.

In similar fashion, the following expression is obtained for the longitudinal propagation mode:

$$T = R^4\sqrt{\rho}\cdot\frac{\sqrt{4\pi}}{H_0a^3}\int_{\varphi_1}^{\varphi_2}\frac{\cos\varphi\,d\varphi}{\sqrt{[(\cos^2\varphi-\cos^2\varphi_0)(1+3\sin^2\varphi)]}}$$

where the integral is an elliptical type. The values of this integral for the six stations are given in Table 2, column 3. In both expressions for T, we determined the value of the parameter $R^4\sqrt{\rho}$ that best fitted the experimental data. Table 1, column 5, gives the computed times of travel for the slow mode, and Table 2, column 5, gives the computed times for the fast mode.

The remarkable agreement between theoretical and experimental values, as evidenced by a comparison between columns 2 and 5 of Table 1, and between columns 2 and 5 of Table 2, seems to be a strong empirical indication of the existence of hydromagnetic wave ducts. The existence of such ducts would also explain better the observed intensity and sharpness of the signals received.

By the use of careful compilation of recent observations of ion density as a function of height in the upper atmosphere[4], and current estimates of the composition of the upper atmosphere, we computed the empirical function $R^4\sqrt{\rho}$ versus height, and found that our values for $R^4\sqrt{\rho}$ for the slow and fast modes corresponded to heights of 1,700 km. and 2,500 km. respectively, for the two ducts.

The refraction mechanism needed to bend most of the high-altitude hydromagnetic energy, produced by the detonation, into a very high-altitude duct has not yet been studied in great detail. Also, the correct mathematical relation between wave normal and propagation direction for our case of concentric ducts in an anisotropic medium has not yet been worked out. However, from the well-known formula for the radius of curvature of a beam being propagated in an inhomogenous medium, $r = v / dv/ds$ (where ds is a line element vertical to the direction of propagation), it follows (for longitudinal hydromagnetic waves) that the beam will be bent into a circular path around the Earth at a height where the ion density changes in proportion to R^{-8}. By using the empirical ion density versus height data, we find that this phenomenon occurs at a height about midway between 2,000 and 3,000 km., which independently supports the duct hypothesis.

A more detailed paper is being prepared for publication.

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[1] Berthold, W., Harris, A. K., and Hope, H., *J. Geophys. Research* (in the press).

[2] Berthold, W., Harris, A. K., and Hope, H., "World-Wide Effects of Hydromagnetic Waves due to *Argus*", manuscript in preparation, U.S. Army Signal Research and Development Laboratory, Fort Monmouth, New Jersey, 1959.

[3] Newman, P., *J. Geophys. Research*, 64, 923 (1959).

[4] Townsend, J. W., *Science*, 129, 80 (1959).

GEOPHYSICS

Tektites and the Earth

The glassy tektites found strewn over the surface of the Earth have long been a scientific enigma. Many hypotheses have been produced to account for their origin and many writers seem to favour an origin beyond the atmosphere, in inter-planetary space. At this juncture it is well to consider several factors that argue against an extra-terrestrial origin.

The bulk constituents of tektites are the same as the constituents of certain sedimentary rocks[1], particularly sandy clays and shales; the twisted silica particles are almost certainly fused grains of sand; tektites are always found associated with sediments. These are arguments in support of the thesis that tektites result from a fusion of the sedimentary rocks of the Earth.

There are notable discrepancies in the percentage of minor constituents of tektites and sediments, but this may be attributed to leaching and weathering processes acting on the sediment before the tektite was formed. The minor constituents that are not subject to differential weathering all support a terrestrial origin. For example, the ratio of nickel to cobalt in tektites is 2·5, in close agreement with sediments[1], while meteorite material invariably shows a ratio between 6 and 60. The deuterium/hydrogen ratio[1] is 0·015 mole per cent, which agrees with the ratio found in the natural waters of the Earth and is considerably higher than the value accepted for non-terrestrial objects. The ratio between strontium-87 in tektites to the total is 0·0702. This value does not correspond to the values for stony meteorites, but agrees closely with the strontium-87 content of sea water and marine sediments[1]. A measurement of the ratio of lead isotopes again shows agreement with terrestrial rocks, and the uranium-thorium-lead balance[1] is more characteristic of Cainozoic sediments than any other material that the astronomer or geologist would expect to find in the entire solar system.

One of the most significant ratios for meteorites is that of iron to nickel, which is usually close to 20 and seldom, if ever, greater than 100. Unfortunately the ratio of iron to nickel on the Earth varies over wide limits for the various types of rock, and is probably affected by differential leaching and weathering in sedimentary deposits. Despite these uncertainties it is of interest to consider the iron-nickel ratio. Tektites show a ratio between 500 and 3,000[1,2], which is considerably higher than that of meteorites and corresponds to the average sediment. Collisions in space are responsible for the dispersal of nickel-iron fragments in stony meteorites and even the surface of the Moon must be contaminated with the 20 : 1 iron-nickel ratio. Thus one would have expected a higher percentage of nickel if tektites were connected in any way with an extra-terrestrial object, whether it be the Moon, an asteroid or meteorite.

Radioactive dating shows that the age of tektites agrees fairly closely with the age of the sedimentary rocks in the tektite area[1]. That is to say, the tektites were probably molten fragments at some instant during the deposition of the sediments. Cosmic-ray bombardment produces helium, neon, argon and other spallation isotopes in the meteorites, which give a measure of the duration of exposure of the specimen in space. So far no measurement has been able to show that the exposure time of tektites is different from zero, and we must conclude that after the melting process tektites were screened from cosmic rays, presumably by the atmosphere of the earth.

These facts offer convincing evidence that tektites were formed on the Earth's surface from sandy sediments by the momentary application of intense and localized heat. I consider that it is more prudent to assume as a working hypothesis that tektites are earthy objects and consider extra-terrestrial origins only when the hypothesis fails.

The suggestion that tektites were produced by the collision of meteorites or comets with the Earth is consistent with a terrestrial origin. The energy of impact would be sufficient to melt the country rock within and around the crater. Impact theories do not adequately account for the widespread nature of tektite fields such as those in Australia and the East

Indies. Furthermore, the original craters have never been found, although we recognize other craters several million years old, nor do tektites contain iron spherules as are found in the glass near recognized meteor craters.

Fusion by lightning has been discounted by many authors[1] because lightning usually produces a long tubular fulgurite whereas tektites are solid and sometimes show pronounced stream-lining or 'flight form'. Fulgurites formed in sand are certainly tubular, but the action of lightning on other sediments, such as a sandy clay, is not known. Recently clay fulgurites, formed in Kingston, Ontario, were brought to my attention by Wallace H. Robb. The objects were partially fused ellipsoids ranging in size from an inch to several inches. We hope to make further studies on these objects. It is perhaps possible for the buttons with flanges, dumbbells and other shapes to be formed by fusion within the ground. It is not correct to assume that these shapes can only be produced by the melting of a tektite during supersonic passage through the air. The usual flight form of meteorites is an irregular cone, the material being blown away as fast as it melts. Meteorites never show flanged buttons, dumbbells, etc.

At Boston University we have commenced a study of tektites, with the support of a N.A.S.A. research grant, and we are considering the hypothesis that tektites are produced by lightning acting on sandy sediments. Lightning offers a ready explanation for the chemical and isotope composition of tektites, their poor annealing and fused silica particles, their age and their widespread yet infrequent occurrence. Artificial tektites have already been produced[2] by the action of a high-energy electron beam on the sandy clay found, on a recent field trip[4], to be associated with the tektites in Texas. Further experiments will continue in an attempt to reproduce the 'flight forms' by fusion, abrasion and etching.

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November 2.

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[1] Symposium on Tektites, *Geochim. et Cosmochim. Acta*, 14 (1958).

[2] Plunkett, J. D., Hawkins, G. S., and Wolfson, S. H. (unpublished work at O. Hommel Co., Pittsburgh, Pa.).

[3] McCaig, J. B., Hawkins, G. S., and Wolfson, S. H. (unpublished work at High Vacuum Equipment Corp., Hingham, Mass.)

[4] Wolfson, S. H. (unpublished work).

[See also p. 291 of this issue.—EDITORS.]

PHYSICS

Time of Residence of Radioactive Debris in the Stratosphere

THE world-wide distribution of strontium-90 from nuclear weapons tests has been extensively studied, especially by the University of Chicago Sunshine Project and others. It has been established that the cumulative deposition has a strong peak in the middle latitudes of the northern hemisphere, and the rate of deposition has repeatedly shown a maximum in the spring.

Libby[1] assumed originally that the debris injected into the stratosphere has a mean time of residence of 10 ± 5 yr. and will be distributed rather uniformly over the globe. He ascribes the peak in the middle latitudes and the increase in deposition in spring to the tropospheric part of the debris.

Based on extensive measurements, Stewart *et al.*[2] have maintained that at places remote from testing areas nearly all strontium fallout is of stratospheric origin. They propose a selective downward mixing of stratospheric debris at middle latitudes, which could explain both the peak and the increase in the spring. Martell[3], however, submits an alternative view; namely, that these phenomena are due largely to recent tests of intermediate- and high-yield weapons which have injected debris into lower and intermediate levels of the stratosphere, debris which has a relatively short time of residence in the stratosphere.

Libby and others[4] now also evaluate the time of residence for stratospheric debris at middle latitudes as considerably shorter, perhaps 2–4 yr.

We have for some time made determinations of the ratio, *r*, of the amounts of strontium-89 and strontium-90 on monthly samples of precipitation from a number of stations in Denmark, including the Faeroes and Greenland. The location of the stations, given to the nearest degree, are as follows:

		Longitude	Latitude
B	Oestermarie, Bornholm	15 E.	55 N.
C	Copenhagen, Zeeland	13 E.	56 N.
T	Thorshavn, The Faeroes	7 W.	62 N.
G	Groennedal, Greenland	48 W.	61 N.
S	Soendre Stroemfjord, Greenland	50 W.	67 N.

Our results are given in Fig. 1. The accuracy of the measurements varies with the strength of the samples as shown.

It can be seen that the ratio *r* in August and September 1958 is around 15. As part of the fallout may be tropospheric, $r = 15$ is an upper value for debris from the stratosphere at that time. The ratio increases in October for some (the northernmost) stations to a little less than 40. From February 1959 it again decreases for all stations. The two dotted lines are drawn with a slope corresponding to the decay of strontium-89.

We believe that the two results for Copenhagen in October and November indicate that very little dust from explosions in October reach Copenhagen before December.

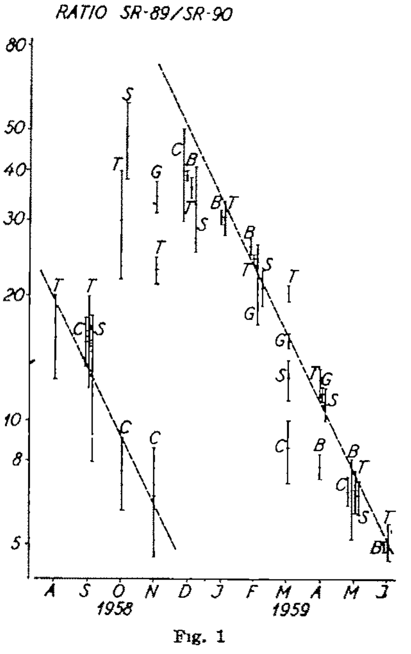

Fig. 1

The results for the period January–June follow the slope of the lines remarkably well. Combined with the cessation of weapon tests around November 1, this seems to show that for a period of five–six months starting two–three months after the last injections in the stratosphere, and for 5 stations distributed nearly along a line 2000 miles long, the composition of the fallout corrected for the decay is very uniform.

Since the time of residence in the troposphere is much shorter and stratospheric debris from older tests would give a lower value of *r*, it is concluded that a considerable part of the fallout in the spring of 1959 over a large area was stratospheric, but originated from the tests in northern latitudes in the autumn of 1958.

From the increase in *r* as given by the vertical distance between the two lines in Fig. 1, it is inferred that 40–50 per cent of the strontium-90 fallout in the period February–June comes from these tests.

These measurements are part of work carried out for the Danish Defence Research Board, and I thank the Board for permission to publish these results.

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Sept. 22.

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[1] Libby, F. W., *Proc. U.S. Nat. Acad. Sci.*, **45**, 738 (1957).

[2] Stewart, N. C, Osmond, R. G. D, Crooks, R. N., and Fisher, E M., *AERE HP/R* 2354 (Oct. 1957).

[3] Martell, E. A., *Science*, **129**, 1197 (1959).

[4] *Science*, **129**, 884 (1959).

Measurement of Ion Beam Currents using a Hall Effect Magnetometer

Ion and electron beam currents are normally measured in terms of either a collected current or the beam power dissipated in a calorimeter. Direct-current collection is complicated by secondary emission at the collector surface, and calorimeters are difficult to use when the beam power is low. For both methods the beam must be interrupted in order to measure the current. The technique described here avoids these disadvantages by measuring the magnetic field associated with the particle beam.

The magnetic field-strength in the vicinity of a beam of charged particles is given by $H = i/5r$, where i is the net current in amperes and r the radial distance in cm. from the beam axis. If $i = 1$ m.amp. and $r = 1$ cm. then $H = 2 \times 10^{-4}$ oersted. Fields of this intensity have been detected using the Hall effect in indium antimonide[1]. If the magnetic field generated by the beam is concentrated on the indium antimonide crystal then currents smaller than one milliampere may be detected.

A 'Mumetal' rod 1·3 cm. in diameter was bent into a 6·0-cm. diameter ring and annealed. An indium antimonide crystal 0·025 cm. thick was placed in a 0·040-cm. air gap built into the ring. This indium antimonide magnetometer was used in conjunction with a transistor oscillator and amplifier. In order to demonstrate the principle the ring assembly was mounted in a vacuum chamber through which passed a 1·0-cm. diameter electron beam of 1 keV. energy. Two current balancing loops, as shown in Fig. 1, were used to cancel the magnetic field due to the beam. The magnetometer was then used as a null indicating device and at the null point the beam current equalled the current through the balance loops.

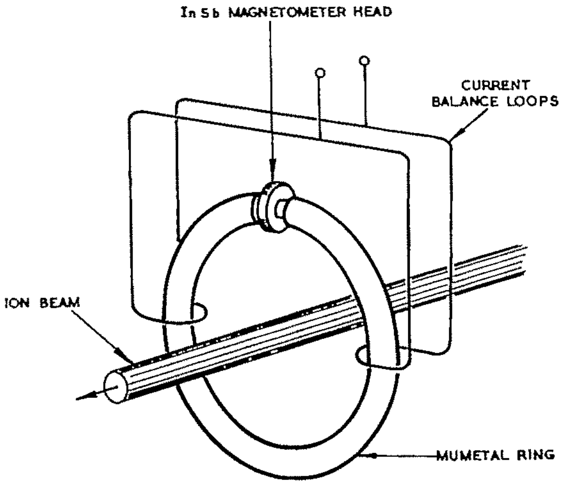

Fig. 1. Schematic diagram of magnetometer ring assembly

Two methods of applying the magnetometer were investigated. In the first method a steady electron beam was directed through the ring with the crystal excited by a 1 kc./s. alternating current. A balance was obtained by adjusting the current through the balance loops until the value of the 1 kc./s. Hall voltage equalled that for zero beam current. The smallest beam current detectable above the noise-level was about 10 μamp. Although a balance was easily obtained for larger currents the accuracy of the device was mainly limited by the zero drift caused by small changes in temperature of the crystal unit. Displacement of the beam from the centre of the ring did not appear to alter the balance condition.

In the second method the crystal was excited by a steady direct current while the beam was modulated at 1 kc./s. Only the alternating component of the Hall voltage was fed into the amplifier. In this case zero drift was virtually eliminated and the smallest detectable current was slightly less than 10 μamp.

The technique offers a new experimental approach to certain problems involving ion beam current measurement. Ionic processes may be studied without the complications associated with conventional detectors.

We are grateful to Miss P. Dunmow and Mr. N. A. C. Thompson for their help in the experiments. Permission to publish has been granted by the Admiralty.

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[1] Hilsum, C., Proc. 1958 Brussels Conf. Solid State.

Cavity Formation in Simple Pipes due to Rupture of the Water Column

When the water-hammer surges that accompany rapid velocity changes in pipes are of such a kind as to produce a fall to vapour pressure, cavities are formed. Theory and analysis at present available rest largely on empirical evidence of changes in pressure alone. In the case of rupture, a recent publication appears to support methods of analysis already in use[1], and suggests cavity formation would take place at the control (gate) end of a straight horizontal pipe.

Studies of the changes in pressure at the centre of a 60-ft. horizontal length of 1-in. copper tube as well as near the control end suggest a new but logical development of the normal theory. The usual principle of the independence of the direct and reflected waves expressed in the basic wave equations can no longer be tacitly accepted, and any such waves which may meet along a pipe must be algebraically summed to ascertain whether the resultant pressure tends to yield a value less than or equal to vapour pressure. It is believed that such a tendency will be invariably found to exist during all rupture periods that occur with the exception of the first. Because pressures less than vapour pressure cannot be tolerated, additional cavity formation is necessary to ensure compatibility of pressure and velocity changes.

Fig. 1 illustrates the result of the theoretical analysis proposed for a specific example. Additional cavity formation occurs at point *D*, due to the simultaneous arrival at this point of a normal reflected wave and a new direct wave occasioned by the resurge of the water column at *A* at the end of the first rupture phase.

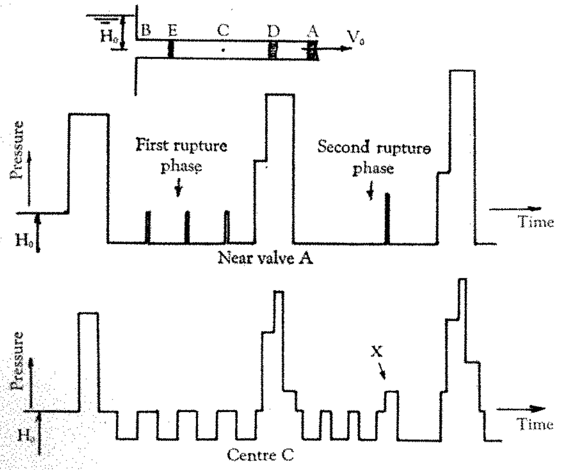

Fig. 1. Graphical solution for rupture

The cavity at *D* in the example shown disappears before the normal cavity at *A*, and evidence of this is seen in the positive pressure rise (marked *X*) at *C* during the second rupture phase. In the case shown further peculiarities of the pressure variations are due to the formation of a complementary cavity at *E*.

Pressure records support the theory, and also observations of a transparent pipe with photographic aid confirm the formation of the cavity as predicted.

It is considered that analytical and experimental investigations of this cavity formation will enable: (a) The phenomenon of rupture of the water column to be explained. (b) The growth and decay of cavities under the action of estimable forces to be described, including the effects of entrained air. (c) The assumed mathematical relationship between pressure and velocity changes to be studied, in view of the fact that the moving cavity serves as an index of water velocities that exist during certain phases.

Work which has been in progress for some time is proceeding along these lines.

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[1] Gayed, Y. K., and Kamel, M. Y. M., *Proc. Inst. Mech. Eng.*, advance copy 34/58.

METALLURGY

A Model of Metal Fatigue

The important part played by the disturbance of the surface of a fatigue specimen by to-and-fro slip movement in the slip bands has been emphasized by much recent work, especially that of W. A. Wood[1]. If the slip in alternate cycles were exactly reversed in the same atomic planes, then there would be no surface disturbance. On the other hand, extrusions or intrusions will be formed if by some mechanism such as that proposed by Mott[2] the dislocations which have moved in one direction along a certain path return by a different one, so as to go round a closed circuit in the same sense at each cycle. There is another possibility: that the dislocations make their return journeys along paths which are shifted in a random fashion with respect to the previous paths. If the shifts are comparable with the width of the slip band, this is equivalent to assuming that the slip is distributed at random at each half-cycle among the available slip planes, and that the distribution of slip at each half-cycle is independent of the distribution of slip in previous cycles.

This random distribution of slip will roughen the surface, each part of which is engaged in a random walk along the slip vector. This roughening will lead to a re-distribution of the stress, so that in later cycles the slip will tend to be concentrated in the valleys already formed on the surface. The subsequent surface movements will make some of the valleys less deep, but some will become deeper, and on these the slip will be even more concentrated. I have set up a rough model of this process in mathematical form, in order to see whether valleys deep enough to lead to cracks could be formed in a reasonable number of fatigue cycles.

These calculations will be published elsewhere. The main result is that after t cycles the fraction f of the valleys of width w which have depths between z and $z + dz$ is given by:

$$f = F \exp - [\{\sqrt{(z + w)} - \sqrt{w}\}^2 / b\,\varepsilon\,t]$$

where F is a slowly varying function of z, w and t. The slip vector b and the plastic strain ε in the slip band are taken as constant. The distribution consists of a Gaussian peak near $z = 0$ and an exponential tail at large z governed by the factor $\exp - z/b\varepsilon t$.

If we suppose that at some large value of z, say Z, the valleys become deep enough to form cracks, we can regard $Z/b\varepsilon$ as a time constant T controlling the rate at which valleys attain the depth Z, and therefore the life-time of the specimen. The first valleys will attain a depth Z after approximately $T/15$ cycles.

It remains to make a rough estimate of the numerical magnitudes involved. We suppose that a valley with a depth ten times its width can be regarded as an incipient crack. Thus $Z \sim 10w$, giving $T \sim 10w/b\varepsilon$. If we insert as typical values, $w = 10^{-4}$ cm., $b = 5 \times 10^{-7}$ cm. (corresponding to the passage of a group of about 20 dislocations at each slip), and $\varepsilon = 10^{-3}$, we get for T about 2×10^6 cycles. The first incipient cracks will appear at about $1{\cdot}3 \times 10^5$ cycles. The satisfactory agreement with the order of magnitude found in practice for life-times suggests that it would be worth while to investigate a more realistic model of this type.

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[1] Wood, W. A., Int. Conf. on Fatigue of Metals, 1956. (Institute of Mechanical Engineers, London), p. 531. *Phil. Mag.* (viii) **3**, 692 (1958), and papers cited there.

[2] Mott, N. F., *Acta Metallurgica*, **6**, 195 (1958).

Oxidation of Titanium–Iron Alloys

Above the transition temperature, the oxidation of titanium proceeds by a layer of titanium oxide (TiO_2) growing outwards by the migration of anion vacancies. Concurrently, a solid solution layer of alpha-titanium containing dissolved oxygen grows inwards. The alpha-layer is fairly uniform in thickness over the surface of the specimen. Micro-hardness measurements show a very marked hardness gradient across it, indicating an oxygen concentration gradient and suggesting that the rate of growth of the alpha-layer is controlled by the diffusion inwards of interstitial oxygen.

It has been found that the addition of small amounts of iron to titanium, while not markedly affecting the overall rate, modifies the structure of the alpha-layer. Alpha-plates form rapidly along some of the beta grain boundaries, plates about 0·5 mm. long forming in about 15 min. at 875°C. Later, the alpha-plates thicken slowly, and a two-phase alpha/beta layer of roughly uniform thickness is formed around the specimen and grows inwards. There is no hardness gradient within the alpha-plates, suggesting that oxygen diffusion is not the controlling step in their formation.

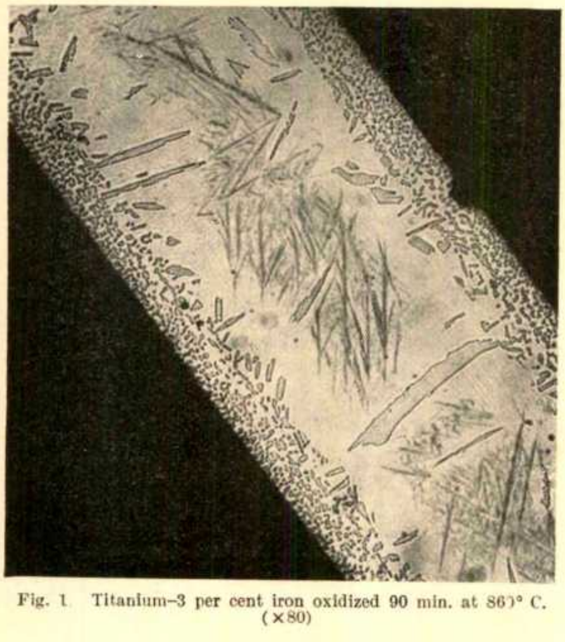

Fig. 1 Titanium–3 per cent iron oxidized 90 min. at 860° C. (×80)

However, when a specimen containing 3 per cent iron is quenched after oxidation, only the beta-phase remote from the alpha-plates is transformed martensitically, as is shown in Fig. 1. Duwez[1] has shown that above 4 per cent iron the martensite start temperature is less than room temperature and beta-phase is retained. From this it is deduced that the iron content in the beta-phase adjacent to the alpha-plates is greater than 4 per cent. Alpha titanium does not dissolve iron to an appreciable extent[2] and it would appear that the iron ejected from the advancing alpha-plates raises the concentration of iron significantly. It is not possible to determine the variations in the iron content of the beta-phase by micro-hardness measurements, because in the neighbourhood of 4 per cent iron the hardness of quenched titanium varies very little with iron content[3]. The oxygen content, as indicated by the hardness of the alpha-phase[4], is about 2 per cent by weight at 850°C. and 2·5–3 at 900°C. which, if local equilibrium is assumed, indicates an iron content of about 6–7 per cent by weight[5].

It is concluded, therefore, that there is an iron gradient in the beta-phase from the original 3 per cent at the centre of the grains remote from the alpha-plates to about 6 per cent by weight at an alpha/beta interface. The rate of thickening of the alpha-plates, and the rate of thickening of the alpha/beta two-phase layer at the specimen surface, are apparently determined by the rate of diffusion of iron away from the interface.

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Sept. 9.

[1] Duwez, P., *Trans. Amer. Soc. Metals*, **45**, 934 (1953).

[2] Hansen, M., McPherson, D. J., and Rostoker, W., "Constitution of Titanium Alloy Systems", U.S. Dept. Comm. Office of Tech. Services P.D. 111508 (1953).

[3] Worner, A. W., *J. Inst. Met.*, **80**, 213 (1951).

[4] Jenkins, A. E., and Worner, A. W., *J. Inst. Met.*, **80**, 165 (1951).

[5] Allen, N. P., Schofield, T. H., and Mellish, B., *J. Inst. Met.*, **82**, 536 (1953).

RADIOCHEMISTRY

Preparation of Radiochemically Pure Tritium Nuclear-labelled Morphine

Morphine has been labelled with carbon-14 in the N-methyl position by chemical synthesis[1,2] and randomly by biosynthesis[3,4]. Because of the metabolic lability of the N-methyl group and the need for considerable quantities of a product of high specific activity, studies on the preparation of tritium-nuclear labelled morphine were initiated. The Wilzbach procedure[5] of exposure to tritium gas was utilized for the introduction of the tracer element into the morphine molecule. The preparation of biologically stable tritium-labelled morphine by this technique had been described previously[6], and the radiochemical purity of the product was reported. We have found, however, that the exposure to tritium gas of morphine base or morphine sulphate in finely divided form, and subsequent purification to remove the labile tritium and radio-decomposition products, uniformly gave a seemingly pure crystalline morphine which showed two distinct areas of radioactivity when paper chromatograms were developed using correctly buffered paper and the proper solvent mixtures. One area contained a greater proportion of radioactivity (approximately 65–70 per cent of total) but negligible mass of material (not detectable by spray reagents or by visualization with ultra-violet light, less than 1 per cent). Similarly when normorphine base or its hydrochloride was tritiated in the same manner and purified, the morphine synthesized from it by the method of Andersen and Woods[1] showed the same trace radio contaminant. When paper chromatograms were prepared from the above tritium-labelled 'morphine' using non-buffered paper and several solvent mixtures, that is, *n*-butanol, formic acid, and water (12 : 1 : 7 v/v); 2·3 per cent *p*-toluene sulphonic acid in *n*-butanol and water (100 : 14-v/v), etc., all the radioactivity was located in one area. This area was the alkaloidal spot as localized by spraying with iodoplatinate alkaloidal reagent or by visualization of the shadow with ultra-violet light. The R_F values were identical with parallel non-labelled morphine.

Paper chromatograms obtained with paper buffered at *p*H values varying between 5·6 to 8·0 (Table 1) and the solvent systems of *tert*-amyl alcohol and water or *tert*-amyl alcohol, *n*-butyl ether and water (80 : 7 : 13 v/v)[7], effected a separation of the radioactive impurity from morphine. By the use of co-chromatography with non-radioactive dihydromorphine, the paper chromatograms gave presumptive evidence that the radiochemical impurity in the tritium-labelled 'morphine' was tritium-labelled dihydromorphine. Separation of the two areas of radioactivity was not possible when the paper was buffered at higher *p*H values, namely, 9·25.

Crude tritium-labelled normorphine hydrochloride was converted to free base, crystallized from methanol and the product reprecipitated from an acetic acid solution by dilute ammonia. (The exposure was performed by the New England Nuclear Corp., Boston, Mass.) The normorphine was converted to morphine by the method of Andersen and Woods[1]. The specific activity (128 μc./mgm.) of the resulting morphine did not change on three serial crystallizations. The amount of the trace radiocontaminant remained unchanged or was only partially removed by vacuum sublimation of the 'chemically pure morphine', or fractional crystallization of its diacetyl derivative followed by the conversion of acetylated product back to morphine. Attempts to remove the tritium-labelled dihydromorphine with basic or acid-washed alumina columns, or cellulose columns, were not successful.

Table 1. TYPICAL R_F* VALUES OF COLD AND RADIOACTIVE MORPHINE AND DIHYDROMORPHINE CALCULATED FROM CHROMATOGRAMS OBTAINED WITH WHATMAN No. 1 PAPER

Compound	*p*H of buffer solution applied to paper				
	5·7^a	7·0^b	7·2^b	7·9^b	9·25^c
Morphine	0·56	0·28	0·37	0·66	0·82
Tritium-labelled morphine	0·55	0·29	0·36	0·65	
Dihydromorphine (non-labelled)	0·41	0·15	0·18	0·54	0·82
Tritium-labelled 'dihydromorphine'	0·40	0·17	0·15	0·56	
Mixture of known non-labelled dihydromorphine and eluted area of tritium labelled 'dihydromorphine'	0·40				

The paper strips were dipped in the 0·2 *M* buffer solution, allowed to drip dry in a hood for 1–2 hr., placed in a glass cylinder containing the solvent mixture for 12–14 hr. (overnight) and the chromatogram then developed with the selected solvent mixture. The scanning of dried chromatogram strips was done by eluting 1-cm. strips with absolute methanol, evaporation in counting vials and subsequent counting in a Packard Tricarb liquid scintillation spectrometer.

* The values are not meant to be absolute and may vary somewhat with temperature, nature of buffer used or extent of saturation of paper in chamber. Non-labelled compounds were run as references in all experiments. The temperature was 23·5° ± 0·5° C. The values for radioactive compounds were calculated from the centre of maximum radioactivity.

[a] *Tert*-amyl alcohol, saturated with 1 : 1 (v/v), 0·2 *M* acetate buffer (this buffer was also used to saturate the paper).

[b] *Tert*-amyl alcohol, *n*-butyl ether and water (80 : 7 : 13 v/v) and paper buffered with 0·2 *M* phosphate buffer.

[c] 0·2 *M* borate buffer; solvent mixture as in footnote *b*.

Radiochemically pure morphine was separated by descending chromatography on Whatman 3 *MM* paper (5 mgm./large sheet) buffered with 0·2 *M* phosphate buffer, *p*H 7·0, and developed with a *tert*-amyl alcohol, *n*-butyl ether and water (80 : 7 : 13) system. The morphine band located under ultra-violet light was extracted with absolute methanol in a Soxhlet extractor.

The methanol extract was mixed with cold carrier and evaporated to dryness under nitrogen and reduced pressure. The morphine was separated from this residue and passed over a small neutral activated alumina[8] column to give pure morphine base. The specific activity was about 5–10 μc./mgm. with a one to three isotope dilution with cold carrier. The same procedure using cold dihydromorphine as carrier gave radiochemically pure dihydromorphine.

Morphine exposed to tritium by the Wilzbach procedure shows addition at the 7 : 8 double bond giving the dihydro-radiocontaminant having a negligible amount of total mass but a greater fraction of total radioactivity. Since this material is not biologically identical with morphine, it is essential to remove the former from morphine to be studied biologically. A detailed report will appear elsewhere.

We acknowledge the support of U.S. Public Health Service Grant No. *B*–625 and Michigan Memorial Phœnix Project Grant No. 101, University of Michigan.

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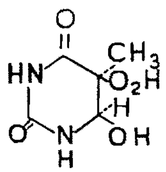
[1] Andersen, K. S., and Woods, L. A., *J. Org. Chem.*, **24**, 274 (1959).
[2] Rapoport, H., Lovell, C. H., and Tolbert, B. M., *J. Amer. Chem. Soc.*, **73**, 5900 (1951).
[3] Battersby, A. R., and Harper, B. J. T., *Chem. and Ind.*, 363 (1958). Leete, E., *ibid.*, 977 (1958).
[4] Achor, L. B., and Geiling, E. M. K., *Anal. Chem.*, **26**, 1061 (1954).
[5] Wilzbach, K. E., *J. Amer. Chem. Soc.*, **79**, 1013 (1957).
[6] Achor, L. B., *J. Pharmacol. Exp. Therap.*, **122**, 1A (1958).
[7] Brossi, A., Haflinger, O., and Schnider, O., *Arzneim. Forsch.*, **5**, 62 (1955).
[8] Reichstein, T., and Shoppee, C. W., *Discuss. Farad. Soc.*, **7**, 305 (1949).

RADIATION CHEMISTRY

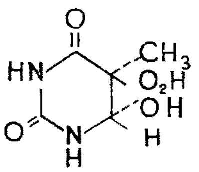
Organic Hydroxy-Hydroperoxides: a Class of Hydroperoxides formed under the Influence of Ionizing Radiations

IN continuation of our previous work on nucleic acids and related compounds[1], it has now been found that many substances, when irradiated with, for example, γ-rays, X-rays, in aqueous solution in the presence of oxygen, are capable of forming hydroxy-hydroperoxy compounds, many of which were previously unknown.

Compounds belonging to this class which may be of interest from a biological point of view are the hydroxy-hydroperoxides, which are formed from different pyrimidine bases, for example, thymine and uracil. Among these, the thymine hydroxy-hydroperoxide is rather stable and can be isolated from the irradiated solutions. In support of an earlier suggestion[2], it has now been found that the peroxides produced by radiation are the (*cis*) and (*trans*)-4-hydroxy-5-hydroperoxide (I) and (II):

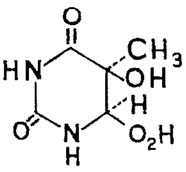


(I)

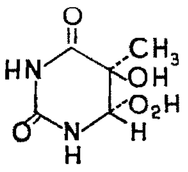


(II)

It is of interest to note that these compounds are not decomposed by catalase. The isomeric 5-hydroxy-4-hydroperoxide should also be possible in a *cis* and *trans* form, so that in addition to (I) and (II) the following compounds may exist:



(III)



(IV)

All these compounds have in fact now been made from thymine by chemical methods by Dr. B. Ekert[3] at the Pasteur Institute. They can be separated, for example, by paper chromatography by means of the solvent system *n*-propanol (80 parts), *N* hydrochloric acid (15 parts) on Whatman No. 1 paper and can also be characterized in this way.

Another convenient way of characterizing these different hydroperoxides was found by measuring the rate constants for the liberation of iodine in the reaction of the hydroperoxide with iodide in aqueous solution. The second-order rate constants for the different hydroxy-hydroperoxides from thymine are given in Table 1:

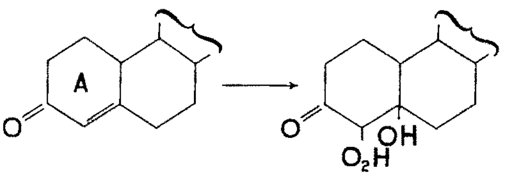
Table 1. RATE CONSTANTS FOR THE REACTION OF DIFFERENT HYDROPEROXIDES WITH IODIDE AT *p*H 4 0 AND 25° C. (Measurements carried out by Mr. J. F. Ward)

Compound	Rate constant (l. mol.$^{-1}$ sec.$^{-1}$)
Hydroperoxide by radiation from thymine	1·59
4-Hydroxy,5-hydroperoxy thymine (I, II)	1·60
4-Hydroperoxy,5-hydroxy thymine (III, IV)	7·58
Hydrogen peroxide	1·18 × 10^{-2}

The rate constant for hydrogen peroxide, measured under the same conditions, has also been included: a comparison of the values shows that the thymine hydroperoxides are very much more reactive than hydrogen peroxide. The rate constant of the compound produced by radiation is practically identical with that of the 4-hydroxy,5-hydroperoxy isomer. The 4-hydroperoxy,5-hydroxy compounds (III, IV) are, under the conditions in question, nearly five times more reactive than the isomer with the hydroperoxy group in the 5-position. On the other hand, we could not detect any appreciable differences between the corresponding *cis* and *trans* isomers. The reason for this may be that either the differences are too small, or in solution a relatively rapid *cis–trans* isomerization takes place.

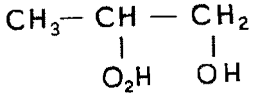
Uracil also forms a corresponding hydroxy-hydroperoxide which is of comparable stability; on the other hand, the hydroperoxide from cytosine appears to be much less stable in solution.

Another new group of hydroxy-hydroperoxides has been obtained from various steroids by means of ionizing radiations[4]. Peroxides derived from steroids have recently again attracted some interest as possible sources of the carcinogenic effects of such substances. The compounds described by Fieser *et al.*[5] and Schenck *et al.*[6] are ordinary hydroperoxides. The study of the action of ionizing radiation on aqueous solutions of, for example, deoxycorticosterone, cortisone and related steroids has shown that these and similar steroids are capable of forming hydroxy-hydroperoxides. Although these compounds have not yet been isolated in a pure form, their chemical and spectroscopic behaviour shows clearly that the OH and HO_2 attach themselves to the double bond of ring *A*, namely:



and/or the corresponding isomer with the OH and HO_2 groups exchanged, which may also be formed to a lesser or greater extent. There are, of course, again two possible stereoisomers for each structure. This type of structure is supported also by the disappearance of the ultra-violet absorption associated with the chromophoric α,β unsaturated ketone grouping in ring *A*. In the case of deoxycorticosterone, which has been studied in some detail, it has been found that two different hydroperoxides are formed in the ratio of approximately 2 : 1. In addition to the hydroperoxide on ring *A*, a second one, which is associated with the side-chain, is also formed, as was shown by a corresponding decrease in the tetrazolium blue reaction.

With regard to some simpler olefinic molecules, it may be of interest to note that propylene forms a hydroxy-hydroperoxide in a comparatively high yield ($G = 2{\cdot}6$ mol./100 eV.) when irradiated in aqueous solutions in the presence of oxygen. The structure of this hydroperoxide was found to be (Clay, P. G., and Whiston, J., unpublished results):



$$\begin{array}{ccccc} CH_3- & CH & - & CH_2 \\ & | & & | \\ & O_2H & & OH \end{array}$$

It can thus be stated that, in general, the action of ionizing radiations on ethylenic systems in the presence of oxygen can give rise to more or less stable hydroxy-hydroperoxides. These compounds may be of considerable biological interest, and bearing in mind the important and unique role of oxygen in connexion with the radiosensitivity of living cells, further study of the biological effects of such compounds is desirable.

We thank Dr. B. Ekert for supplying us with samples of the synthetic thymine hydroperoxides and the Rockefeller Foundation for financial support.

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[1] Daniels, M., Scholes, G., Weiss, J., and Wheeler, C. M., *Nature*, 178, 157 (1956); *J. Chem. Soc.*, 226 (1957).

[2] Weiss, J., in "Organic Peroxides in Radiobiology", 42 (edit. by Haissinsky, M.) (Masson, Paris, 1958). Cf. also Okada, S., *ibid.*, 46.

[3] Ekert, B., and Monier, R., *Nature*, 184, B.A.58 (1959).

[4] Oakey, R. E., Ph.D. thesis, Durham (1958).

[5] Fieser, L. F., Greene, Th. W., Bischoff, F., Lopez, G., and Rupp, J. J., *J. Amer. Chem. Soc.*, 77, 2928 (1955).

[6] Schenck, G. O., Gollnick, and Neumuller, O. A., *Ann. Chem.*, 603, 46 (1957). Schenck, G. O., and Neumuller. O. A., *ibid.*, 618, 194, 202 (1958).

Electron Paramagnetic Resonance Observations on Irradiated Thiols and Disulphides

THE unique role of sulphur in radiation damage is amply borne out by recent electron paramagnetic resonance work[1–3]. In a current investigation on the mechanism of action of the sulphur-containing radioprotective compounds, quantitative electron paramagnetic resonance studies have been carried out on a series of irradiated thiols and disulphides.

The substances were irradiated in the polycrystalline state at room temperature with conventional X-rays (220 keV.). Irradiations were carried out in vacuum as well as in air. In most cases, the radiation doses were less than 5×10^5 r. The spectrometer used operates at a frequency of 9,200 Mc. per sec. The spectra obtained represent the first derivative of the actual absorption curves. Quantitative results were obtained by double integration and comparison with measurements on standard systems. The more significant observations may be summarized as follows:

Dose-effect relationships. The qualitative electron paramagnetic resonance spectra exhibited in some instances a continuous and distinct change with increasing radiation dose, even in the comparatively low dose-range used (Fig. 1). Moreover, for all compounds studied the number of resonance centres detected did not increase linearly with the radiation dose, as exemplified in Fig. 2. This dependence of the yield on the radiation dose may in part explain the large discrepancies found in the literature concerning the number of electron volts needed to produce free radicals[4–6].

Time effects. The number of detectable paramagnetic resonance centres showed an unexpected relationship to the time elapsed after the irradiation. For some compounds (for example, cystine, penicillamine) the number of free radicals N seemed to decay in vacuum according to the general equation: $N = a \ln (t) + b$, where t is the time and the parameters a and b vary with the substances. In the case of other compounds (for example, homocystine, cystamine), the number of resonance centres increased by approximately 10–20 per cent during the first hours after the irradiation. In these instances the peak was reached in 3–4 hr. and the number then decreased, the decay obeying the general formula stated above. The quantitative changes were associated with qualitative changes in the electron paramagnetic resonance spectra in the case of homocystine and cystamine. The spectra of cystine and penicillamine, on the other hand, were constant with time.

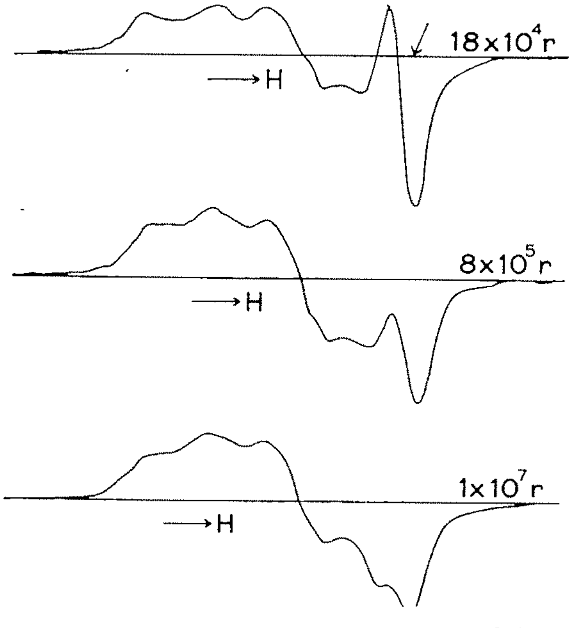

Fig. 1. The effect of the radiation dose on the qualitative electron paramagnetic resonance spectra of cystamine dihydrochloride irradiated in vacuum. The measurements were carried out 30 min. after the conclusion of the exposure. The arrow indicates the position of g-factor of free electron spin. The total width of the spectrum is about 150 gauss

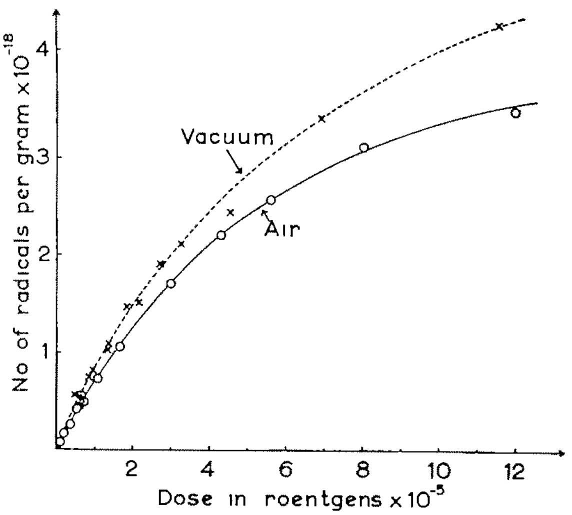

Fig. 2. The relationship between the radiation dose and the number of paramagnetic resonance centres induced in cystamine dihydrochloride. From the curves, 'initial yields' (calculated from the straight line portion of the curves) were found to be 7 and 8 eV. per induced spin in vacuum and in air, respectively

The observed increase in the number of resonance centres in the case of homocystine and cystamine suggests the possibility that several types of free radicals are initially formed, one of which may decay with the formation of two or more other types. The concurrent qualitative spectral changes may be taken to support this view.

Structural effects. In contrast with previous views[1], structural factors were found to exert a pronounced effect on the qualitative electron paramagnetic resonance spectra of thiols and disulphides (Fig. 3). Whereas cysteamine, cysteine, and cystine gave closely similar resonance patterns, marked differences were obtained when the carbon chain was extended (for example, 1-mercapto-7-aminoheptane, homocystine) or branched (for example, penicillamine). Substitutions in the amino group (N,N′-tetramethylcystamine, N-morpholyl-cysteamine, N-piperidylcysteamine) likewise resulted in pronounced changes in the resonance patterns. In general, these chemical alterations introduced a hyperfine structure in the high-field part of the spectra.

The above-mentioned findings bear out the fact that electron paramagnetic resonance measurements are influenced by several parameters which must be clearly defined in qualitative as well as in quantitative electron paramagnetic resonance studies. They also demonstrate that the chemistry of the radiation of thiols and disulphides in the dry state is more com-

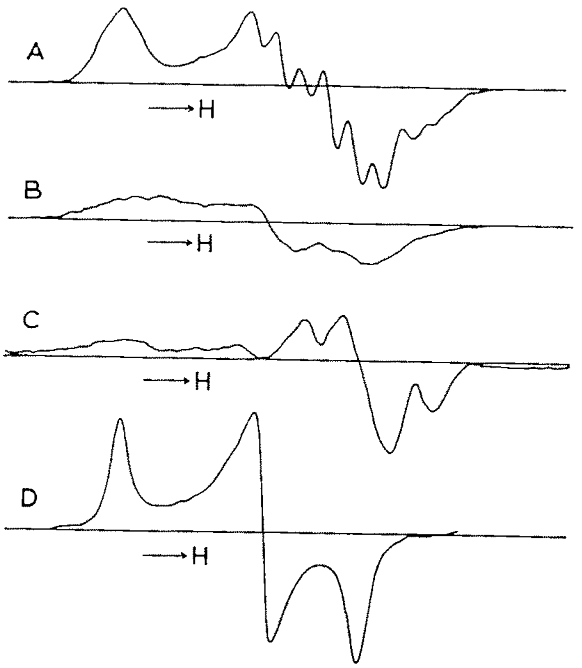

Fig. 3. Examples of variations in the qualitative electron paramagnetic resonance spectra with variations in the chemical structure. Dose, 5×10^4 r. Otherwise, conditions as in Fig. 1. *A*, N,N′-tetramethylcystamine dihydrochloride; *B*, *l*-cysteine hydrochloride; *C*, reduced glutathione; *D*, *d*-penicillamine. In the course of 2–3 days, the spectrum of glutathione changed to resemble closely that of cysteine

plex than previously recognized. The distinct differences observed between the spectra of various thiols and disulphides open up the possibility of studying the chemical properties of irradiated mixed disulphides, a question which is of interest from a radiation protection point of view[7,8]. Such studies are in progress.

The details of this work will be published elsewhere.

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[1] Shields, H., and Gordy, W., *J. Phys. Chem.*, 62, 789 (1958).

[2] Gordy, W., and Shields, H., *Rad. Res.*, 9, 611 (1958).

[3] Smaller, B., and Avery, E. C., *Nature*, 183, 539 (1959).

[4] Zimmer, K. G., Ehrenberg, L., and Ehrenberg, A., *Strahlentherapie*, 103, 3 (1957).

[5] Box, H. C., and Freund, H. G., *Nucleonics*, 17, 66 (1959).

[6] Randolph, M. L., and Parrish, D. L., *Rad. Res*, 9, 170 (1958).

[7] Eldjarn, L., and Pihl, A., in "Progress in Radiobiology", ed. by Mitchell, J. S., Holmes, B. E, and Smith, C. L. (Oliver and Boyd, London, 1956).

[8] Eldjarn, L., and Pihl, A., in "Mechanism in Radiobiology", ed. by Forssberg, A., and Errera, M. (Academic Press, Inc., New York, 1960; in the press)

CHEMISTRY

Experimental Test of Verwey and Overbeek's Theory

Ghosh and Rakshit[1] have measured the ζ-potentials of a titanic oxide sol in the presence of equicoagulating concentrations of electrolytes. An attempt has been made here to calculate the ζ-potentials of the colloid particles of a titanic oxide sol at flocculating concentrations of electrolytes from an analysis of the nature of the curves of total potential energy of interaction of two spherical colloid particles, and to compare these calculated values with those obtained by Ghosh and Rakshit by electro-osmotic measurements.

The total potential energy V is calculated from the following expression:

$$V = V_R + V_A \qquad (1)$$

where V_R and V_A are the repulsive and attractive potential energies of interaction of two colloid particles respectively.

Derjaguin[2] has shown that for small surface potential ψ_0 and large values of $\varkappa a$ the repulsive energy V_R of interaction between two colloid particles, each of radius a, can be calculated from the following expression:

$$V_R = \frac{Da\psi_0^2}{2} \ln \left[1 + \exp\{-\tau(S-2)\}\right] \qquad (2)$$

where D is the dieletric constant and τ and S are equal to $\varkappa a$ and R/a respectively, where R is the distance between the centres of two colloid particles.

Hamaker's[3] expression for the attractive energy V_A of interection of two spherical particles can be written in the following form:

$$V_A = -\frac{A}{6}\left[\frac{2}{S^2-4} + \frac{2}{S^2} + \ln\frac{S^2-4}{S^2}\right] \qquad (3)$$

where A is the Van der Waals–London constant.

In order to calculate the values of V from equations 1, 2 and 3 it is necessary to know the values of $\varkappa$, a and A. The values of $\varkappa$ have been calculated from the following expression:

$$\varkappa = \sqrt{\frac{4\pi\varepsilon^2 \Sigma n_i z_i^2}{DkT}} \qquad (4)$$

where ε is the electronic charge, D the dielectric constant of the medium, k the Boltzmann constant, n_i the number of ions of the ith type per c.c. of the solution and z_i is its valency. Taking $T = 35°$ C. and $D = 75$, the values of $\varkappa$ at flocculating concentrations of electrolytes are calculated from equation 4 for a titanic oxide sol[1] and given in column 3 of Table 1.

Table 1

Titanic oxide sol

Electrolyte	Concentration of electrolyte ($N \times 10^3$)	$\varkappa \times 10^{-6}$	ζ observed (mV.)	ζ calculated (mV.)
$K_4[Fe(CN)_6]$	2 86	0·8847	10 3	13·94
Na_3-citrate	2 72	0 7716	9 0	12·93
Na_2SO_4	4·5	0 8596	12 8	13 72
NaCl	103 00	3 357	28 2	28·2
$K_4[Fe(CN)_6]$ + NaCl	1 43 51·50	2 456	25·0	23 97
Na_3-citrate + NaCl	1 33 51 5	2 435	24·8	23 85
Na_2SO_4 + NaCl	2 237 51 5	2 45	24·2	23·95
Na_2SO_4 + NaCl	4 114 5 15	1 114	16·1	15·86

Since the colloid particles increase in size rapidly before they finally flocculate, the value of a at the point of flocculation is very high and has been arbitrarily taken to be equal to 1×10^{-4} cm. for this calculation. It can also be shown that the calculation of the ζ-potential is not appreciably affected by even a large variation of a from this value.

The value of A has been evaluated from an analysis of the potential energy curves. Identifying ψ_0 with ζ and taking ζ = 28·2 mV., $\varkappa = 3{\cdot}357 \times 10^6$ and $a = 1 \times 10^{-4}$ cm. for sodium chloride, the values of V

for different S values are calculated from equations 1, 2 and 3 for a particular value of A. A curve is drawn by plotting the values of V against S. Different curves are obtained for different values of A. According to Verwey and Overbeek[4] the transition between stable and flocculated systems is represented by the potential curve with a weak potential barrier for which the top coincides with the horizontal axis. Therefore the value of A for which the maximum of the potential energy curve just touches the S-axis represents the value of A for this particular sol. The value of A evaluated from the graph is found to be $3{\cdot}884 \times 10^{-13}$. In the evaluation of A by the above method it has been assumed that the measured ζ-potential of the coagulum in presence of 0·0103 N sodium chloride represents the true ζ-potential of the colloid particles.

After determining the value of A by the above method the ζ-potential of the flocculated particles for any other electrolyte is determined by drawing similar potential energy curves. Knowing the value of κ from equation 4 and taking $A = 3{\cdot}884 \times 10^{-13}$ and $a = 1 \times 10^{-4}$ cm. a total potential energy curve is drawn for a particular value of ζ. Different curves are obtained for different values of ζ. The value of ζ for which the maximum of the total potential energy curve just touches the S-axis represents the ζ value for this particular electrolyte. The ζ values evaluated by this process for different electrolytes are given in column 5 of Table 1.

The ζ values evaluated from the potential energy curves are found to be very close to the ζ values obtained by electro-osmotic measurements. The above agreement between the experimental and calculated ζ values give direct support to Verwey and Overbeek's theory.

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[1] Ghosh and Rakshit, *Sci. Culture*, 18, 498 (1953).

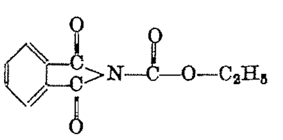
[2] Derjaguin, *Trans. Farad. Soc.*, 35, 203 (1940).

[3] Hamaker, *Physica*, 1058 (1937).

[4] Verwey and Overbeek, "Theory of the Stability of Lyophobic Colloids", p 108 (Elsevier. Amsterdam, 1948).

Synthesis of Phthaloyl Amino-Acids under Mild Conditions

THE synthesis of phthaloyl amino-acids, using amino-acids as starting material, often proceeds under rather drastic conditions[1]. Phthaloyl amino-acid esters, however, can be prepared under less drastic conditions[2–4]



We found that N-carboethoxy phthalimide is an excellent reagent for the preparation of phthaloyl amino-acids under mild conditions. The reaction, which is carried out in aqueous solution at room temperature, applies to many functional groups

The reagent is easily prepared from potassium phthalimide and ethyl chlorocarbonate in dimethyl formamide or from phthalimide, triethylamine and ethyl chlorocarbonate in the same solvent. It is easily crystallized from ethanol or isopropyl alcohol.

To 30 ml. of water (temp. 17°–20°) 1·5 gm. of glycine (0·02 mole), 5·75 gm. sodium carbonate 10 aq. (0·02 mole) and 4·5 gm. of N-carboethoxy phthalimide were added. The mixture was stirred for about 15 min. The solution was then filtered and the filtrate acidified. After crystallization from water and drying, 3·72 gm. of phthaloyl glycine (yield: 90·5 per cent) m.p. 191° were obtained.

With only small modifications of the above-mentioned procedure, the compounds presented in Table 1 were prepared

Table 1

Compound	Yield	m p
Phthaloyl-β-alanine	91 per cent	152°
Phthaloyl-L-glutamic acid	65 per cent	160°
Phthaloyl-DL-serine	95 per cent	152°
Phthaloyl-L-asparagine	85 per cent	199°
Phthaloyl-DL-phenylalanine	90 per cent	178°
ε-phthaloyl-L-lysine*	85 per cent	232°
Phthaloyl-DL-methionine	96 per cent	102°
Diphthaloyl-L-cystine	92 per cent	120°
Phthaloyl-L-leucine	92 per cent	110°

* Prepared from the copper complex of lysine

Phthaloyl-L-glutamic acid prepared by this method has the same rotation as reported by Clark-Lewis and Fruton[5]. After removing the phthaloyl groups in diphthaloyl-L-cystine, the cystine thus obtained (without further purification) in a yield of 85 per cent has the same rotation as our starting material, 208° in 1 N hydrochloric acid.

From these facts we conclude that the introduction of the phthaloyl group by our method respects the optical activity of the amino-acids.

Details and proposed mechanism will be published elsewhere (*Rec. Trav. Chim.*).

I would like to thank G. I. Tesser for the benefit of many discussions.

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[1] Billman, J H., and Harting, W F , *J. Amer Chem Soc.*, 70, 1473 (1948)

[2] Bose, A K., Greer, F., and Price, C C , *J. Org. Chem.*, 23, 1335 (1958)

[3] King, F. E., and Kidd, D A. A., *J Chem. Soc*., 3315 (1949).

[4] Balenovic, K., Gaspert, B , and Stimac, N , *Croat Chem. Acta*, 29, 93 (1957).

[5] Clark-Lewis, J. W , and Fruton, J. S , *J. Biol. Chem* , 207, 477 (1954)

Paper Chromatography of Acidic Carbohydrates

CATIONIC detergents, such as cetyltrimethylammonium bromide ('Cetavlon') and cetylpyridinium chloride, have been used extensively in recent years for the fractionation of polysaccharide mixtures[1–4]. The quaternary ammonium cations present in these detergents form salts with acidic polysaccharides which often have solubility properties quite different from those of the usual metallic salts; for example[1], cetyltrimethylammonium chondroitin sulphate is soluble in aliphatic alcohols up to n-pentanol and is insoluble in water. Many purification procedures have utilized the water-insolubility of these quaternary ammonium polysaccharide salts, but surprisingly, no serious attempt seems to have been made to exploit their solubility in organic solvents. This communication describes some preliminary experiments directed towards developing convenient methods for the separation of acidic carbohydrates, making use of the solubility of their cetylpyridinium salts in organic solvents. Current interest in these laboratories in the purification and identification of sugar sulphates, suggested that it would be useful if this property could be utilized in a method for the paper chromatography of these substances. It has been found that when a small amount of detergent is incorporated into the mobile (organic) phase of certain solvent systems for paper chromatography, the R_F values for sulphated derivatives are increased by a factor of about ten, and some useful separations can be achieved. Two such systems are:

(*a*) *Butanol/ethanol/water* (3 : 1 : 1 *by vol.*; 100 *ml.*) *plus cetylpyridinium chloride* (3 *gm.*). The R_F values for some sugar sulphates in this solvent system are given in Table 1. The mixture obtained by the direct sulphation of glucose (with pyridine–sulphur trioxide) was resolved into at least five components (Table 1). The major monosulphated component had the same R_F value as glucose-6-sulphate, thus confirming previous results[5]. Small quantities of two other glucose monosulphates were detected, one of which had the same R_F value as glucose-3-sulphate.

Table 1. R_F VALUES OF SOME SUGAR SULPHATES*

	R_F values*	
	Solvent (*a*)†	Solvent (*b*)†
Glucose	0 21	0·18
Glucose-3-sulphate	0 67	0 85
Glucose-6-sulphate	0 41	0 77
Glucose disulphates	1 00‡	1·00‡
Products from direct sulphatation of glucose	0·21, 0 41, 0 55, 0·67, 1·00	0·18, 0·77, 0·85, 1 00

* R_F values are measured relative to the rate of movement of the slower of the two solvent fronts.

† Development period for solvent (*a*) 16 hr.; solvent (*b*) 3 hr

‡ Travels as an extremely compact zone.

(*b*) *Water-saturated methyl ethyl ketone* (100 *ml.*) *plus cetyl pyridinium chloride* (3 *gm.*). Although this solvent system is less useful than system (*a*) for the separation of isomeric glucose monosulphates, it does possess certain advantages. Thus, simple mixtures can be resolved very rapidly; for example, glucose and glucose monosulphate are well separated by a 15-min. development using the ascending technique. (Equilibration of the paper against the solvent vapour prior to development may, however, be advisable when more complex mixtures are to be analysed.) The detection of acidic sugar derivatives in a mixture (for example, a polysaccharide partial hydrolysate) should be rapid and simple since only these substances will move faster than the parent monosaccharide when the detergent is incorporated into the solvent, and slower when it is omitted.

For the location of sugar zones after irrigation of the chromatogram, two methods have been found useful. The silver nitrate–sodium hydroxide reagents[6] can be used if the paper is first rinsed with chloroform to remove the detergent, or *p*-anisidine hydrochloride[7] can be applied directly to the dried chromatogram and, after heating, the spots observed in ultra-violet light.

This work is being continued with the view of developing analogous methods for the paper chromatography of other acidic carbohydrates.

I am grateful to Mr. T. P. Williams and Mr. K. O. Lloyd for the samples of sugar sulphates used in this work, to the Department of Scientific and Industrial Research for financial assistance, and to Prof. Stanley Peat and Dr. J. R. Turvey for their interest and encouragement.

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[1] Bera, B. C., Foster, A. B., and Stacey, M., *J. Chem. Soc.*, 3788 (1955).

[2] Scott, J. E., *Chem. and Indust.*, 165 (1955).

[3] Barker, S. A., Stacey, M., and Zweifel, G., *Chem. and Indust*, 330 (1957).

[4] Palmerstierna, H., Scott, J. E., and Gardell, S., *Acta Chem. Scand.*, 11, 1792 (1957).

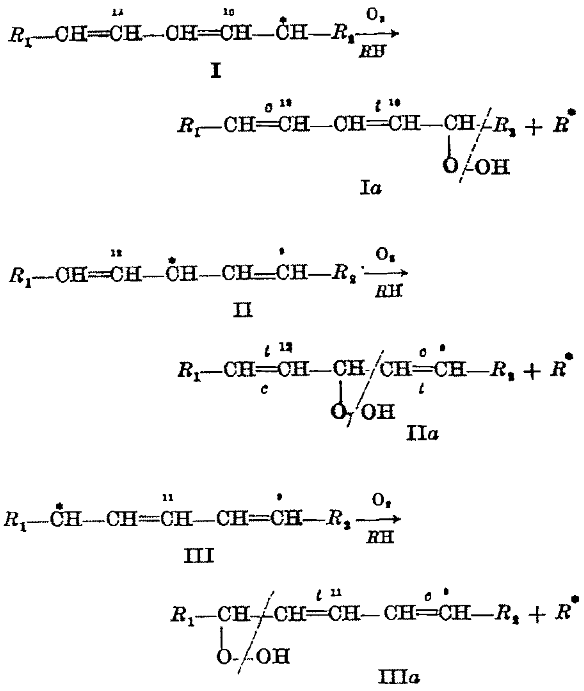
[5] Lloyd, A. G., *Nature*, 183, 109 (1959).

[6] Trevelyan, W. E., Procter, D. P., and Harrison, J. S., *Nature*, 166, 444 (1950).

[7] Hough, L., Jones, J. K. N., and Wadman, W. H., *J. Chem. Soc.*, 1702 (1950).

The Stereo-configuration of 2,4-Decadienals isolated from Oils containing Linoleic Acid

ACCORDING to Farmer's theory (see Bolland[1] and Bateman[2]), the following isomeric hydroperoxides (I*a*, II*a*, and III*a*) are formed in the autoxidation of linoleic acid, from the mesomeric linoleic acid free-radicals (I, II and III):



$$R_1{-}CH{=}\overset{12}{C}H{-}CH{=}\overset{10}{C}H{-}\overset{*}{C}H{-}R_2 \xrightarrow[RH]{O_2} R_1{-}CH\overset{c\ 12}{=}CH{-}CH\overset{t\ 10}{=}CH{-}\underset{O{-}OH}{CH}{-}R_2 + R^*$$

I → I*a*

$$R_1{-}CH{=}\overset{12}{C}H{-}\overset{*}{C}H{-}CH{=}\overset{\cdot}{C}H{-}R_2 \xrightarrow[RH]{O_2} R_1{-}CH\underset{c}{\overset{t\ 12}{=}}CH{-}\underset{O{-}OH}{CH}{-}CH\underset{t}{\overset{c\ \cdot}{=}}CH{-}R_2 + R^*$$

II → II*a*

$$R_1{-}\overset{*}{C}H{-}CH{=}\overset{11}{C}H{-}CH{=}\overset{\cdot}{C}H{-}R_2 \xrightarrow[RH]{O_2} R_1{-}\underset{O{-}OH}{CH}{-}CH\overset{t\ 11}{=}CH{-}CH\overset{c\ \cdot}{=}CH{-}R_2 + R^*$$

III → III*a*

R_1 = $CH_3{-}(CH_2)_4{-}$ RH = linoleic acid molecule
R_2 = $-(CH_2)_7-COOH$ R^* = linoleic acid radical
t = *trans*, *c* = *cis*.

The theoretical stereoconfiguration of the possible isomers has been indicated by Nichols *et al.*[3].

Swift *et al.*[4] have isolated from cottonseed oil (which contains approx. 45 per cent linoleic acid) in various stages of autoxidation 2,4-decadienal, 2-octenal, and hexanal. Recently, Patton *et al.*[5] have published results on the occurrence of 2,4-decadienal in several oils and fats. The formation of these decomposition products can be readily explained by means of the above-mentioned theory. From the

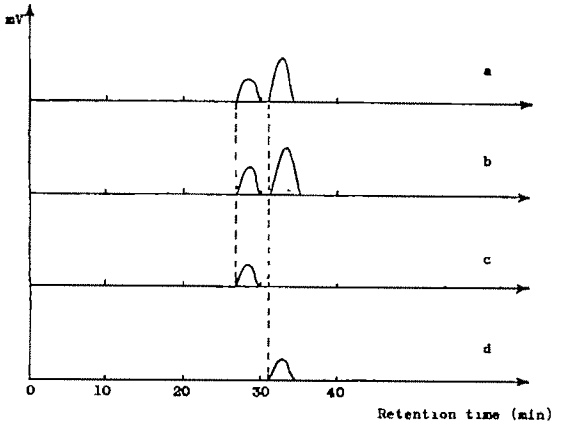


Fig. 1. Gas-liquid chromatography of. *a*, palm oil decomposition product (detail); *b*, groundnut oil, ditto; *c*, synthetic 2(*trans*)-4(*cis*)-decadienal; *d*, synthetic 2(*trans*)-4(*trans*)-decadienal Carrier, 'Celite 545'; immobile phase, silicone oil (30 per cent); mobile phase, nitrogen; pressure, 50 cm. mercury; gas velocity, 50 ml./min.; temperature, 137° C.; length of column, 90 cm.; detector, gas density balance

relevant literature, however, no information can be obtained on the stereo-configuration of the unsaturated aldehydes obtained, notably 2,4-decadienal.

From our investigations with palm oil and groundnut oil it appeared that on subjecting the volatile decomposition products to gas-liquid chromatography[6], two aldehydes could be isolated, which were identified as two isomeric 2,4-decadienals (Figs. 1*a*, *b*; 2*a*, *b*). By partition chromatography of the dinitrophenylhydrazones, following the method of Miss Kramer and van Duin[7], the 2,4-decadienal structure could be established, but not the stereo-configuration.

Two isomeric 2,4-decadienals of known stereo-configuration were synthesized. Using Brown and Subba Rao's method[8] 2(*trans*)-4(*cis*)-decadienal was prepared from 2(*trans*)-4(*cis*)-decadienoic acid. This acid was isolated, in a yield of 3–5 per cent, from *Stillingia* oil, the oil from the seeds of *Sapium sebiferum* Roxb. of which the stereo-configuration has been established by Crombie[9]. By application of Doebner's method (cf. Forss and Hancox[10]) 2(*trans*)-4(*trans*)-decadienal was synthesized, starting from hexanal. As the synthetic products and the two aldehydes isolated from volatile decomposition products gave similar results in the gas-liquid chromatographic analysis (Figs. 1*c*, *d*) and in the infra-red analysis (Figs. 2*c*, *d*), the conclusion is justified that the two natural 2,4-decadienals obtained possess the 2(*trans*)-4(*cis*) and 2(*trans*)-4(*trans*) stereo-configuration. From the gas chromatogram these *trans,cis* and *trans,trans* compounds were found to be present in the ratio of 28 : 72.

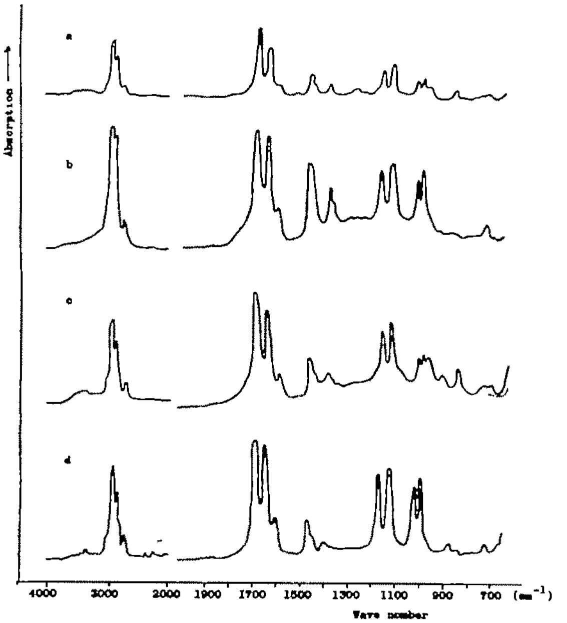


Fig. 2. Infra-red spectra of: *a*, 2(*trans*)-4(*cis*)-decadienal from palm oil (first peak in Fig. 1*a*); *b*, 2(*trans*)-4(*trans*)-decadienal from palm oil (second peak in Fig. 1*a*); *c*, synthetic 2(*trans*)-4(*cis*)-decadienal (cf. Fig. 1*c*); *d*, synthetic 2(*trans*)-4(*trans*)-decadienal (cf. Fig. 1*d*).
To record these spectra, a 'Unicam' 100, double-beam infra-red spectrophotometer was employed (rock-salt prism, microplate technique)

The above affords new, indirect proof that the (I*a*) isomer of linoleic acid hydroperoxide is formed as one of the primary autoxidation products as found spectrophotometrically by Sephton and Sutton[11]. Then it must be assumed that isomer (I*a*) serves as precursor for 2(*trans*)-4(*cis*)-decadienal. The more stable 2(*trans*)-4(*trans*)-decadienal can be formed, either as a secondary reaction product during further rearrangement of isomer (I*a*) to 9-hydroperoxy-10(*trans*)-12(*trans*)-decadienoic acid, or as a result of the instability of 2(*trans*)-4(*cis*)-decadienal. More detailed results will be published elsewhere.

We wish to thank Mr. J. H. Recourt for his assistance in carrying out the gas-liquid chromatography experiments. Further, our thanks are due to Unilever Research Department, Port Sunlight, for supplying the sample of *Stillingia* fatty acids and to the management of Unilever N.V., Rotterdam, for permission to publish these data.

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[1] Bolland, J. L., *Quart. Rev.*, 3, 1 (1949).
[2] Bateman, L., *Quart. Rev.*, 8, 147 (1954).
[3] Nichols, jun., P. L., *et al.*, *J. Amer. Chem. Soc.*, 73, 247 (1951).
[4] Swift, C. E., *et al.*, *J. Amer. Oil Chemists' Soc.*, 26, 297 (1949).
[5] Patton, S., *et al.*, *J. Amer. Oil Chemists' Soc.*, 36, 280 (1959).
[6] Beerthuis, R. K., and Keppler, J. G., *Nature*, 179, 731 (1957).
[7] Kramer, P. J. G., and van Duin, H., *Rec. trav. chim.*, 73, 63 (1954).
[8] Brown, H. C., and Subba Rao, B. C., *J. Amer. Chem. Soc.*, 80, 5377 (1958).
[9] Crombie, L., *J. Chem. Soc.*, 1007 (1955).
[10] Forss, D. A., and Hancox, N. C., *Austral. J. Chem.*, 9, 420 (1956).
[11] Sephton, H. H., and Sutton, D. A., *J. Amer. Oil Chemists' Soc.*, 33, 263 (1956).

Effect of Flow-Rate and Sample-Size on Column Efficiency in Gas-Liquid Chromatography

THERE have been a number of investigations as to the effect of carrier gas flow-rate on separation and column efficiency in gas-liquid chromatographic systems[1–4]. Many of the results present contradictory pictures of the effect exerted by this parameter. Keulemans[1] states, "for a certain set of operational conditions there is an optimum linear gas velocity, but the optimum is very flat and the gas rate is consequently not a highly critical parameter". James and Martin[2] have shown that column efficiency is sensitive to flow-rate and that a decrease in flow-rate increases column efficiency. Their results show a

27 per cent increase in column efficiency when the flow-rate was decreased from 24 to 10 ml./min. Bethea and Wheelock[3] reported that column efficiency, as measured by the H.E.T.P., is approximately tripled by raising the flow-rate from 5 to 27·5 ml./min. They found that increasing the flow-rate beyond the latter point gave only meagre increases in efficiency.

Mellor[4] found the critical flow-rate to be independent of temperature, pressure, column length, and load size, but dependent on column diameter and particle size of the column packing support.

During an investigation of the separation of two terpene hydrocarbons, α-terpinene and D-limonene, by means of gas-liquid chromatography, it was of interest to determine whether better separation could be achieved by selection of an optimum carrier gas flow-rate and sample size. It is a well-established fact that the efficiency of separation in a gas-liquid chromatographic column improves as the size of the sample is reduced[1,2]. Thus, with low column-load and optimum gas-flow, a high efficiency should be achieved.

The apparatus employed to separate α-terpinene from D-limonene was an 'Aerograph' model *A*-90-*C* (manufactured by Wilkins Instrument and Research, Inc., Walnut Creek, California). The column was constructed of stainless steel tubing, ¼ in. outside diameter and 10 ft. in length. The support material used was 'Sil-O-Cel' *C*-22 diatomaceous earth fire-brick (30–60 mesh). Fractions of this were sieved to size and further graded by sedimentation in water, after which they were dried at 150° for 16 hr. The liquid phase was introduced by deposition from benzene solution, the packing then being heated in a stream of helium at 150° for 16 hr.

The column was contained in an air thermostat; the carrier gas flow (helium) was controlled by a precision reducing valve and measured by a soap bubble flow meter situated at the column outlet. Temperature and flow control were sufficiently good to make possible the use of a four-channel, hot-wire katharometer that proved virtually noiseless when connected with a 1 mV. recording potentiometer. Sample injection was made with a microsyringe, and the sample preheater was maintained at 150°.

Separations were carried out with the aid of a stationary liquid phase consisting of *LAC*-4-*R*777 (the succinate polyester of diethylene glycol)[5]. This material was applied to the solid support in the amount of 25 per cent w/w. Column temperature was maintained at 100°, and recorder chart speed was 30 in./hr. The column outlet was maintained at atmospheric pressure.

Flow-rates varied from 20 to 145 ml./min. of helium. Table 1 illustrates the effect of flow-rate and sample-size on column efficiency under the experimental conditions described above. The number of theoretical plates was calculated by means of the equation[5]:

$$\text{Number of theoretical plates} = 16\left[\frac{\text{Retention volume}}{\text{Peak width}}\right]^2$$

As the number of theoretical plates can vary with the nature of the component[6], a calculation was made for both peaks on each chromatogram, and the average of the two values is presented in Table 1. It should be noted that the theoretical plate values for both peaks varied in a manner similar to that displayed by the average of the two values.

With a 0·1-μl. sample load, the theoretical plate value increased 63·8 per cent when the flow-rate of the carrier gas was decreased from 145 to 45 ml./min. An additional decrease in the flow-rate to 20 ml./min. decreased the theoretical plate value 43 per cent. Employing a 0·3-μl. sample load, this same decrease in flow-rate, that is, from 145 to 45 ml./min. increased the theoretical plate value 46·9 per cent. and a further decrease in flow-rate to 20 ml./min. decreased the theoretical plate value 26·3 per cent. At a sample load of 0·5 μl. the theoretical plate value increased 25·8 per cent and then decreased 24·6 per cent in going through the same flow-rate changes. Thus at constant sample size, a plot of flow-rate versus theoretical plate value for these systems shows a pronounced maximum.

When one operates at optimum flow-rate and varies the sample size from 0·1 to 0·3 μl., the theoretical plate value decreases by 12·5 per cent. Increasing the sample size to 0·5 μl. reduces the theoretical plate value by 39·5 per cent.

Bethea and Wheelock[3] have noted that as the flow-rate is increased the optimum sample size for maximum column efficiency increases. It may be noted that the optimum flow-rate appears to shift toward 60 ml./min. at the higher column load (Table 1).

Table 1

	Flow-rate (ml./min.)	Theoretical plates		
Sample size		0·1 μl.	0·3 μl.	0·5 μl.
	20	1499	1508	1096
	30	1967	1751	1167
	45	2144	1905	1282
	60	2040	1851	1366
	[illegible]	—	—	[illegible]
	92	1592	1601	1188
	145	1309	1297	1086

Investigations of the effects of these operational parameters are being continued and will be published in the future.

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[1] Keulemans, A. I. M., "Gas Chromatography", 23 (Reinhold Publishing Corporation, New York, 1957).

[2] James, A. T., and Martin, A. J. P., *Biochem. J.*, 50, 679 (1952).

[3] Bethea, R. M., and Wheelock, T. D., Abstracts of the 2nd Biannual International Gas Chromatography Symposium, 1 (published by the Instrument Society of America, Pittsburgh, Penn., 1959).

[4] Mellor, N., in Desty, D. H. (ed.), "Vapour Phase Chromatography", 67 (Butterworth Scientific Publications, London, 1957).

[5] Lipsky, S. R., and Landowne, R. A., *Ann. N.Y. Acad. Sci.*, 72 (13), 666 (1959).

[6] Johnson, H. W., and Stross, F. H., *Anal. Chem.*, 30, 1586 (1958).

Cathode-Ray Presentation of Chromatograms

FROM the results of investigations on nylon capillary columns[1], it became apparent that there were two possible applications for the capillary system. The columns could be used to produce very high efficiencies with elution times of 10–60 min., or the high efficiencies could be sacrificed and chromatograms developed in a matter of seconds, with moderate efficiencies of perhaps 5,000 or 10,000 plates. Under these conditions it was found that a chromatogram of a mixture boiling over a range of about 100 deg. C. would be developed in 60 sec., the early peaks being eluted during a period of about

0·5 sec., but it was necessary to use a high-speed recording system with time constants of the order of milliseconds. Such a performance cannot be achieved by the normal potentiometric recorder, and the oscilloscope is an obvious alternative method of presenting the chromatogram.

In addition to the improved speed of response, an oscilloscope is also cheaper to manufacture. The complete chromatographic equipment, incorporating the column, detector and ancillary equipment, could be manufactured for less than the price of a single recorder.

With such an apparatus, it would be possible to analyse a volatile mixture in a matter of seconds, and it could be used as a laboratory instrument for routine analysis. By use of an automatic injection system synchronized with the scan, it would be possible to monitor the product from a plant every minute. Another interesting application would be its use in the investigation of reaction kinetics, where the complete analysis of a volatile mixture in a matter of seconds would be a distinct advantage.

The construction of a suitable display system is comparatively easy. The results shown below were obtained from a normal television amplifier system and power supply, coupled to a 14-in. magnetically deflected cathode-ray tube coated with a phosphor having a 200-sec. persistence. The micro-argon detector devised by Lovelock was used, in conjunction with a modified impedance converter of extremely low time-response. The scan was obtained by a linear-wound potentiometer driven by a constant-speed motor. The column used was 70 ft. of 0·020 in. diameter nylon tubing coated with a total of 8 mgm. of 'Apiezon Oil *A*' as liquid phase, and was run at an inlet pressure of 9 lb./sq. in. A 100 : 1 diluting system was used on the injection, in order that a sufficiently narrow band of solute could be placed on the column[2]. The total charge on the column was about 2 μgm. and the sensitivity of the detector was of the order of 10^{-11} gm./c.c.

A photograph of the trace obtained from the analysis of a petroleum ether mixture is shown in Fig. 1. The first two peaks, *iso*-butane and *n*-butane, are just resolved, the third high peak is *n*-hexane and the last peak is methyl*cyclo*hexane. The total scan of the tube was 50 sec. The first peak, *iso*-butane, was eluted in 31 sec. measured from the time of injection, and the elution curve itself was developed over a period of 0·5 sec. *n*-Hexane was eluted in 40 sec. and *n*-heptane in 80 sec. The efficiency for *n*-hexane on this chromatogram is between 5,000 and 6,000 plates. This is not obvious because the injection is not shown, as it occurred about two-thirds of the way through the previous scan.

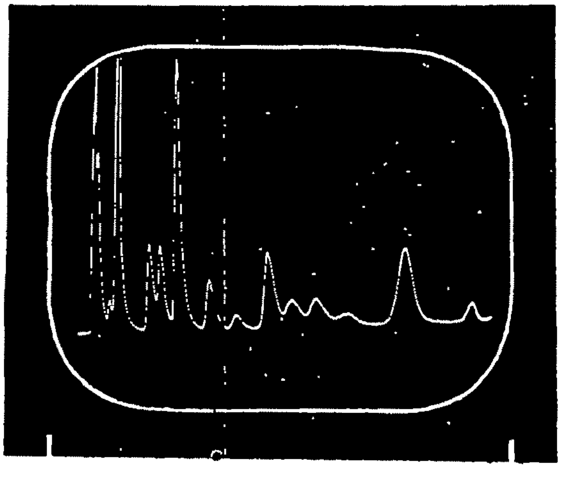

Fig. 1. Chromatogram from cathode-ray tube

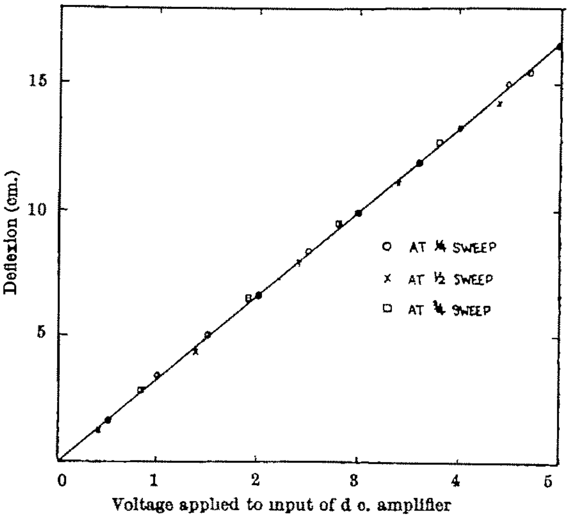

Fig. 2. '*Y* deflexion'/voltage input to d.c. amplifier

The amplifying system was adjusted to give a linear response between voltage input on the d.c. amplifier and centimetres deflexion on the *Y*-axis of the oscilloscope. Thus, providing the micro-argon detector is adjusted to give a linear response, then quantitative results can be obtained from the chromatogram. The relationship between centimetres deflexion and voltage input to the d.c. amplifier is shown in Fig. 2.

The instrument is still under development, and results have shown that higher efficiencies and even shorter times of elution are practical. However, these early results clearly indicate that the oscilloscope system is an alternative method for presenting chromatograms. Full details of this work will be published elsewhere.

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[1] Scott, R. P. W., *Nature*, **183**, 1753 (1959).

[2] Desty, D. H., Goldup, A., and Whyman, B. H. F., "The Potentialities of Coated Capillary Columns for Gas Chromatography in the Petroleum Industry" (to be published).

BIOCHEMISTRY

Metal-Organic Complexes formed during the Treatment of Wood with Metal Salts

A RECENT communication by Belford, Cook and Nevard[1] dealt with the subject of metal-cellulose complexes formed during the treatment of Douglas fir wood with various preservative processes comprising solutions of inorganic salts or mixtures of these. The authors claim that these are formed at room temperatures, and that, in the case of the copper complex, the process by which it is formed differs from the one described by Abrams and Bottoms[2], who have also claimed the formation of a copper-cellulose complex when cellulose-containing materials, such as cotton, are treated with a solution of copper formate followed by heating.

In a study[3] of the process described by Abrams and Bottoms we have been unable to confirm the observations which led them to conclude that a copper-cellulose complex had been formed. Thus, bleached or unbleached cotton fabric treated with copper formate solution according to this method was readily dispersible in a solution of cuprammonium hydroxide of the composition normally used in dispersing textile cellulose for fluidity measurements[4]; the copper in the treated fabric could be readily leached out by dilute ammonia; the decomposition, brought about by heating, of copper formate on the bleached cotton fabric, gave the same colour change when glass fabric was used.

In a study of the kinetics of the thermal decomposition of metallic formates, Kornienko[5] found that the main products of the reaction were a metal oxide and products of the decomposition of formic acid, and that where free metal was present in the reaction products it could be attributed to the reduction of the oxides by the gaseous decomposition products, notably carbon monoxide and hydrogen. It is of interest to note that the mechanism postulated by this author would make it possible for cuprous oxide to be one of the reaction products. While the evidence from X-ray diffraction data for the presence of cuprous oxide in our fabric treated with cupric formate followed by heating is inconclusive, it does not preclude its presence.

Moreover, we found that the resistance to microbiological attack shown by cotton fabric treated with cuprous oxide was similar to that shown by the copper formate treated fabric containing a similar percentage of copper. Thus fabrics containing cupric formate and cuprous oxide at concentrations of 0·43 per cent and 0·26 per cent respectively gave essentially the same resistance in the soil burial test, namely, 6–10 per cent loss of strength for the former and 13 per cent loss for the latter.

In an earlier communication[6] Cook and Nevard refer to the copper formate process for the preservation of wood as described by Walton-Smith *et al.*[7], this being the same process described by Abrams and Bottoms[2]. Here, it is claimed by Walton-Smith *et al.* that a copper-cellulose complex is formed in the wood, and it is stated by Cook and Nevard in the article referred to above that their results prove conclusively the existence of this copper-cellulose complex. However, in the later communication[1], these latter authors give data which, they claim, indicate that the copper-cellulose complex which they believe to have been formed in their experiments is not of the same type as the one postulated by Abrams and Bottoms.

If by 'copper-cellulose complex' Cook and Nevard mean an association of copper and substantially pure cellulose we would take exception to this view, and we would suggest that the complexes formed be regarded as predominantly the product of the interaction of the metal ions with acidic, cation-binding, non-cellulose constituents of the wood. Indeed, in view of the close association of these non-cellulosic materials with the cellulose of wood, it is difficult to envisage the formation of a pure metal-cellulose complex in the amounts claimed by Cook and Nevard under the conditions of their experiments.

It is well known that 'native celluloses' show marked, but varying, tendency to take up metal cations after treatment with acid followed by washing. This effect has been examined by a number of workers and it has been shown that although a highly purified cellulose consisting substantially of α-cellulose has a small cation-binding power, relating probably to the presence of acidic end groups[8], this capacity to bind cations is greatly increased as we approach more nearly to the 'native celluloses' which are known to contain non-cellulosic constituents such as pectins, uronic acids and lignin[9].

In some of their treatments of wood with inorganic salts, Cook and Nevard observed heavy incrustations of electron-opaque material. They also observed that certain fragments of treated wood, whether showing incrustations or not, gave sharp electron diffraction diagrams, characteristic of the treatment; that in the case of copper treatments, these diagrams did not resemble any of the X-ray patterns for the copper salts employed and also that the diffraction diagram faded fairly rapidly with increasing intensity of the electron beam. All these observations are consistent with our view that the complexes formed consist of metal cations bound on the acidic groups of non-cellulosic constituents of the wood.

Such a view is also consistent with the observation of Belford, Cook and Nevard that the reaction leading to the formation of the complexes takes place at room temperature.

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[1] Belford, D. S., Cook, C. D., and Nevard, E. H., *Nature*, 183, 988 (1959).
[2] Abrams, E., and Bottoms, R. R., *Text. Res. J.*, 26, No. 8, 630 (1956).
[3] Rose, Grace R. F., and Bayley, C. H., *Text. Res. J.* (in the press).
[4] Clibbens, D. A., and Little, A. H., *J. Text. Inst.*, 27, T285 (1936).
[5] Kornienko, V. P., *Ukrain. Khim. Zhur.*, 18, 579 (1952); *Chem. Abstr.*, 48, 4945^{4}, (1954).
[6] Belford, D. S., Preston, R. D., Cook, C. D., and Nevard, E. H., *Nature*, 180, 1081 (1957).
[7] Walton-Smith, F. G., Bottoms, R. R., Abrams, E., and Miller, S. M., *Forest Prod. J.*, 6, No. 9, 340 (1956).
[8] Sookne, A. M., and Harris, M., *Text. Res. J.*, 10, No. 10, 405 (1940).
[9] McLean, D. A., and Wooten, L. A., *Indust. Eng. Chem.*, 31, No. 9, 1138 (1939).

Mediation of the Genetic Control of Protein Synthesis in Cell-free Extracts of *Neurospora crassa*

I HAVE recently demonstrated the development of tryptophan synthetase activity by cell-free extracts from conidia of *wild-type Neurospora crassa*. The process appeared to be one of synthesis of enzyme protein, and was associated with the net increase in the protein content of the system[1]. Similar extracts prepared from the *td*$_2$ *tryptophan-less* mutant strain neither contained nor developed any detectable tryptophan synthetase activity, but did catalyse a net synthesis of protein.

The defective component of the mutant extract has been located in the 'soluble' fraction not sedimented by centrifuging at 105,000 *g* for 1 hr. As shown in Table 1, when a mutant 'particle' fraction was supplemented with *wild-type* 'soluble' fraction, considerable tryptophan synthetase activity was developed. Con-

Table 1. LOCATION OF THE GENETIC DEFECT IN EXTRACTS OF *td*$_2$ MUTANT STRAINS

Tryptophan synthetase activities (units/ml. of conidial ext.)

'Soluble' Fraction 'Particle' Fraction	Wild Type Wild Type		Mutant Mutant		Wild Type Mutant		Mutant Wild Type	
Exp. No.	Initial	Final	Initial	Final	Initial	Final	Initial	Final
1	0·21	0·37	0	0	0·21	0·30	0	0
2	0·52	0·72	0	0	0·56	0·79	0	0
3	0·20	0·37	—	0	—	0·30	—	0
4	0·30	0·47	0	0	—	0·44	0	0
5	0·17	0·34	0	0	0·19	0·30	0	0

versely, a mixture of *wild-type* 'particles' with mutant 'soluble' fraction failed to develop any detectable activity.

My method of preparing conidial extracts[1] (see below) could cause contamination by material from the cell nuclei. However, in view of the short duration of the experiments, it seems unlikely that this material contributes significantly to the development of tryptophan synthetase activity observed. Further, extracts prepared from *wild-type* mycelium by milder homogenization procedures also readily form active enzyme (unpublished results). I therefore believe that the defective component of the td_2 mutant extracts is some constituent of the cell 'sap'. Certainly, it would seem that the primary genetically determined defect in the mutant conidium is not located in the 'particulate' fraction corresponding to the microsomes of mammalian cells.

This conclusion has important implications for our understanding of the manner in which complex gene loci control the formation of specific enzymes, and of the mechanism of action of suppressor genes.

Extracts were prepared from conidia harvested from the growth on solid synthetic medium[1]. *Wild-type* cultures were grown at 30° C. for 4 days, and incubated at room temperature (about 20° C.) for a further 4 days. Mutant cultures were grown at 30° C. for 7 days on medium supplemented with 0·04 per cent (w/v) of DL-tryptophan, and incubated a further 7 days at room temperature. The washed and chilled conidia were ground with powdered glass, extracted with cold $M/20$ sodium potassium phosphate buffer (pH 6·5) containing 20 per cent (w/v) sucrose, and separated into 'soluble' and 'small particle' fractions as described in detail elsewhere[1].

The reaction system contained 'soluble' fraction, 'small particle' fraction, 4 per cent (w/v) Difco 'Bacto Casamino Acids', 6·4 mgm. L-cysteine hydrochloride, 50 mgm. hexose diphosphate magnesium salt, 4 mgm. diphosphopyridine nucleotide, 0·4 mgm. calcium chloride, $1{\cdot}26 \times 10^{-1}$ M magnesium sulphate and 42 mgm. of polyvinyl acetate (as a finely dispersed emulsion) in a final volume of 28 ml. In experiment 5 a mixture of 8 mgm. of L-proline plus 3·2 mgm. of each of L-threonine and L-valine replaced half the casein hydrolysate.

Tryptophan synthetase enzyme was precipitated from samples of the reaction system, taken before and after incubation for 1 hr. at 30° C., by a modification[1] of the procedure of Suskind and Kurek[2], and enzymic activity was assayed by a modification[1] of Yanofsky's method[3]. Total protein nitrogen was determined by standard micro-Kjeldahl techniques after precipitation with trichloracetic acid, and extraction of nucleic acids and lipids[1].

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[1] Wainwright, S. D., *Canad. J. Biochem. Physiol.* (in the press).

[2] Suskind, S. R., and Kurek, L. I., *Science*, 126, 1068 (1957).

[3] Yanofsky, C., in "Methods in Enzymology", 2, ed. by S. P. Colowick and N. O. Kaplan (Academic Press, Inc., New York, 1955).

Anærobic Glycolysis of Dispersed Cell Suspensions from Normal and Malignant Tissues

IN studies of cellular metabolism, the potential advantages of using dispersed cell suspensions rather than tissue homogenates or slices have led a number of workers to investigate the methods of preparation and properties of cell suspensions from various tissues[1–5]. Liver parenchymal and kidney epithelial cell suspensions prepared by mechanical disruption of the tissue, with or without prior perfusion of the organ, appear to be morphologically intact when examined by either light or electron microscopy[1,2]. These cell suspensions, however, are apparently metabolically different from tissue slices, since they show negligible endogenous respiration which cannot be restored by glucose or a number of protein and non-protein factors[2,3], and have lost the ability to convert intracellular carbohydrate to lactic acid[3]. The oxidation of succinate by these cells, however, has been reported to proceed at a rate exceeding that observed with slices[2,3], and is independent of exogenous cytochrome c (ref. 4). Liver cell suspensions have also been found to incorporate glycine at a rate seven-fold greater than that observed with homogenates[6].

We have found that cell suspensions from a number of solid and ascitic animal tumours metabolize glucose either aerobically or anærobically, whereas isolated liver and kidney cells do not. This difference in behaviour between these normal and tumour cell suspensions was found to be due to a nearly complete loss of some of the enzymes of the Embden–Meyerhof system from the kidney and liver cells, but not from the tumours tested.

Rat liver and kidney cells were prepared by Anderson's procedure[1], namely, perfusion of the organ with calcium-free Robinson's solution containing 0·027 M citrate, followed by dispersion of the tissue in a loose-fitting homogenizer. The cells were then washed twice with calcium-free Robinson's solution. Cells from solid tumours were prepared in the same fashion, except that the tissues were dispersed without prior perfusion; ascites cells were diluted with the perfusion medium and washed as above. The yield of intact cells varied with source of tissue, but was in close agreement with published results[1]. Suspensions of dispersed cells from the solid Novikoff hepatoma and both the Novikoff and Ehrlich ascites tumours were found to grow at a rate equal to, or better than, untreated tumour mince, or whole ascites fluid, respectively, when transplanted, indicating no loss of viability during isolation.

The lack of respiration of liver and kidney cells in an unfortified system with or without glucose was con-

Table 1 ANÆROBIC GLYCOLYSIS OF CELL SUSPENSIONS

The reaction system (3·0 ml.) contained 7·5 μmoles phosphate (pH 7·4), 20 μmoles magnesium chloride, 49·5 μmoles potassium bicarbonate, and 150 μmoles potassium chloride; 1·0 μmole adenosine diphosphate (ADP), 1·0 μmole diphosphopyridine nucleotide (DPN), and 50 μmoles glucose where indicated. The vessels were gassed with 95 per cent nitrogen–5 per cent carbon dioxide for 10 min. Incubation, 1 hr. at 30° C. The values are μmoles lactic acid/hr./mgm. nitrogen.

Tissue	Minus ADP and DPN Endogenous	Minus ADP and DPN + Glucose	Plus ADP and DPN Endogenous	Plus ADP and DPN + Glucose
Liver	0	0	0	0
Kidney	0	0	0	0
Thymus	0·2	2·0	0·9	2·2
Novikoff hepatoma (solid)	0·3	2·0	0·9	5·0
Murphy lymphosarcoma	0·3	4·4	0·3	4·6
Walker-256	0·3	1·5	0·8	4·6
Gardner lymphosarcoma	0·7	5·3	0·9	8·0
Novikoff hepatoma (ascites)	0·8	4·6	1·0	5·5
Ehrlich ascites	0·5	5·6	0·6	6·0

firmed, whereas tumour cell and thymocyte suspensions show both endogenous and glucose-stimulated respiration. Succinate was oxidized by all cell suspensions studied, and the rate could be stimulated by addition of adenosine diphosphate. Preliminary results of a study of the oxidation of a variety of substrates by these cell suspensions indicate that adenosine diphosphate and diphosphopyridine nucleotide are required for maximal activity.

The results of a comparison of anærobic glycolysis by cell suspensions from various tissues are shown in Table 1. Liver and kidney cells did not catalyse anærobic glycolysis, as measured in terms of lactic acid production/hr./mgm. nitrogen from glucose. There was, however, a small but measurable release of carbon dioxide when measured manometrically. Fortification of the medium with adenosine diphosphate and diphosphopyridine nucleotide did not restore the glycolytic ability of these cells, indicating that the metabolic alteration was not caused by leakage of these co-factors. No activity could be demonstrated using the system described by LePage[7], which was designed to demonstrate glycolysis with homogenates of normal tissues. Thymocytes and all the tumours tested catalysed an active glycolysis, both from endogenous substrates and glucose, which could be stimulated by adenosine diphosphate and diphosphopyridine nucleotide with some tumour cell suspensions.

The inability to produce lactic acid from glucose in liver and kidney cells has been demonstrated to be due to the loss of at least two key enzymes of the glycolytic pathway. In Table 2 the specific activity of aldolase and lactic dehydrogenase of the original dispersed tissue, prior to centrifugation, and the final washed-cell suspension are compared, since measurements of total activity in the fractions are not significant due to variations in the yield of cells from the various tissues. Isolated liver and kidney cells contained negligible aldolase and lactic dehydrogenase, whereas cells from thymus and the tumours retained a significant quantity of both enzymes. The enzymes lost from the normal cells were quantitatively recovered in the wash fluids. Glycerophosphate dehydrogenase has also been found to be completely leached out of the liver and kidney cells. This leakage of enzymes has been demonstrated with liver and kidney cells prepared by other techniques for dispersal[4,5]. Perfusion with various media (for example, serum albumin in saline, whole serum, tissue culture medium 199 and mouse ascitic fluid all containing citrate) did not yield cells which retained these enzymatic activities. Cell suspensions from all solid tumours with the exception of the Murphy lymphosarcoma, which requires drastic treatment for dispersal, retained the greater part of their initial content of these enzymes, as shown by a comparison of the specific activity of the dispersed tissue fraction and the washed cell suspension.

Table 2 ALDOLASE AND LACTIC DEHYDROGENASE ACTIVITY OF TISSUE HOMOGENATES AND CELL SUSPENSIONS

Aldolase activity was determined essentially by the method of Dounce *et al.* (ref. 8). Specific activity equals change in optical density (540 mμ) per mgm. nitrogen of chromogen formed in 15 min. Lactic dehydrogenase determined spectrophotometrically; specific activity is μmoles reduced diphosphopyridine nucleotide oxidized/min./mgm. nitrogen, measured as the initial rate. Values are the average of three experiments

Tissue	Aldolase activity Dispersed tissue*	Aldolase activity Cells	Lactic dehydrogenase Dispersed tissue*	Lactic dehydrogenase Cells
Liver	3·8	0·1	8·6	0
Kidney	3·2	0·1	2·0	0
Thymus	2·2	1·4	3·1	1·7
Novikoff hepatoma (solid)	3·5	2·7	3·5	2·5
Murphy lymphosarcoma	11·7	4·1	3·1	1·8
Walker-256	8·8	4·8	6·0	3·2
Gardner lymphosarcoma	5·2	3·6	3·4	3·6
Novikoff hepatoma (ascites)	6·3†	13·6	4·9†	12·1
Ehrlich ascites	4·2†	4·9	5·4†	8·7

* Tissue after dispersion but prior to centrifugation.
† Total ascites fluid and cells.

Although appearing morphologically intact, liver and kidney cell suspensions have been demonstrated not to be biochemically 'intact' due to the complete loss of some soluble enzymes. The use of such cell suspensions for certain metabolic studies is thus limited. The differential leakage of some enzymes from cell suspensions of malignant tissues and thymus as compared with liver and kidney suggests a fundamental difference in the membrane structure of cells depending on the degree of organization of the tissue source.

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[1] Anderson, N. G., *Science*, **117**, 627 (1953).
[2] Kalant, H., and Young, F. G., *Nature*, **179**, 816 (1957).
[3] Laws, J. O., and Stickland, L. H., *Nature*, **178**, 309 (1956).
[4] Kaltenbach, J. P., *Exp. Cell. Res.*, **7**, 568 (1954).
[5] Branster, M. V., and Morton, R. K., *Nature*, **180**, 1283 (1957).
[6] LePage, G. A., *Cancer Res.*, **13**, 178 (1953).
[7] LePage, G. A., *Cancer Res.*, **10**, 77 (1950).
[8] Dounce, A. L., Barnett, S. R., and Beyer, G. T., *J. Biol. Chem.*, **185**, 769 (1950).

Spectrophotometric Determination of Tryptophan by the Method of Roth and Schuster: Some Sources of Error

IN Roth and Schuster's method[1] for colorimetric determination of tryptophan in vegetable matter, the measurement of the yellow colour is made with a Pulfrich Stufenfotometer with filter *S* 43. The method has often been used, but I have been unable to find any published account of the measurements made spectrophotometrically. I have therefore tested the method for some cereals, etc., by means of a Beckman spectrophotometer, model *D.U.*

It appeared that the absorption curves for the yellow compounds produced by D,L-tryptophan and the vegetable matter, respectively, have the same form only within a limited range of wave-lengths. In preliminary attempts to account for this, it was observed that nitric oxides developed during the process of nitration (especially in the case of substances rich in carbohydrates) are dissolved in the reaction mixture, to which they impart a yellow colour. Furthermore, it was found that lignin, which occurs in lignified vegetable matter, is nitrated, forming vividly yellow compounds, and the determination is thus distorted.

The effect of the nitric oxides was shown by the difficulty of obtaining reproducible determinations. The following experiment showed that the nitric oxides contributed to producing the yellow colour: the reaction mixture was bubbled with nitrogen dioxide at room temperature until the air above the fluid assumed a reddish-brown colour from nitric oxides. Dilutions of the solution showed that as little as 1/16 of the original quantity of nitrogen dioxide gave a measurable extinction at 450 mμ.

The occurrence of nitric oxides in the solutions for analysis can be shown by testing for nitrite (development of a red colour by diazotization of sulphanilic acid and coupling in alkaline fluid with α-naphthol), as nitrous acid develops in the solutions:

$$2\,NO_2 + H_2O \rightleftarrows HNO_2 + HNO_3$$

The nitric oxides could be removed completely by sucking air vigorously through the filtered solutions to be analysed. The procedure only caused a slight change of the extinctions (2 per cent), for which correction can be made.

The solutions for analysis (wheat grain, wheat straw) contained nitrogen dioxide corresponding to $\frac{1}{2}$–$\frac{1}{8}$ of the nitrogen dioxide test solution.

The absorption curves for the nitrated solutions of D,L-tryptophan show a maximum at 330–340 mμ. The extinction curves for the wave-lengths of 380, 400, 420, and 450 mμ (1–10 mgm./50 ml., 1 cm. cuvettes) and 400, 410 . . . 500 mμ (5-cm. cuvettes) were all straight lines.

Samples of grain (wheat, oats, rye, barley) and proteins (gluten, albumen) on nitration yielded yellow products, the absorption curves of which for wave-lengths up to about 440 mμ were steeper than those for tryptophan, while for the range 440–500 mμ they followed those of tryptophan, indicated by the fact that the extinctions here, calculated as tryptophan, yielded the same values.

For hulls of oats and for wheat straw the results were different. On nitration they left orange-coloured precipitates, which could only with difficulty be rinsed free from colour. The solutions showed greater absorption in the range of about 470–500 mμ than tryptophan (crossing of the tryptophan curve). The extinction at 450 mμ, calculated as tryptophan, yielded unusually high values. For wheat-straw (0·5 gm. sample) a tryptophan content of 2·25 per cent was found, corresponding to 25·3 per cent tryptophan in the protein (nitrogen × 6·25).

On going through groups of substances which might be supposed to give rise to the formation of yellow-coloured nitration products, lignin attracted special attention, as this is of an aromatic character and is found in lignified vegetable matter, such as the straw and hulls of grain.

Further evidence was obtained by treatment of samples of lignin (acid-lignin). On nitration, compounds of a vividly yellow colour developed, the absorption curves of which showed the crossing of the tryptophan curve characteristic of wheat straw and oats hulls. At 450 mμ a quantity of 0·01 gm. lignin gave the same extinction (0·102) as 1 mgm. tryptophan (0·105), and Roth and Schuster's method therefore in its present form must be considered unsuitable for vegetable matter with even a slight content of lignin.

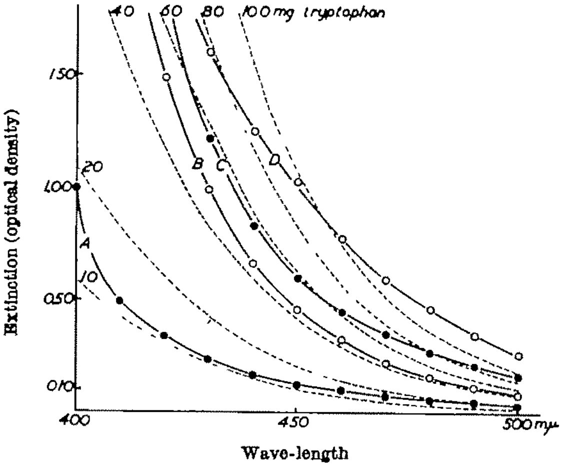

Fig. 1. Absorption curves measured with the Beckman spectrophotometer, model *D U.*, in 5-cm. cuvettes against reaction mixture. Broken curves, D, L tryptophan. Curve *A*, nitrogen dioxide dissolved in reaction mixture; curve *B*, 1·5 gm. wheat grain; curve *C*, 1·0 gm. oats straw; curve *D*, 0 1 gm. lignin. The solutions were diluted to 50 ml. before being measured

As to other kinds of vegetable matter, etc., it is necessary to undertake the measurements at a wave-length higher than about 440 mμ, which necessitates the use of 5-cm. cuvettes. However, it will be desirable to investigate the character of compounds which cause increased absorption below about 440 mμ, as the possibility cannot be precluded that some of the absorption above 440 mμ originates from this.

Roth and Schuster have thoroughly tested a number of amino-acids, etc., with a view to the development of yellow-coloured compounds. The absorption spectra of these have been examined in this Department; a few of them (especially tyrosine, phenylalanine and methionine) showed absorption. The character of the compounds formed is under investigation.

The acid-lignin used has kindly been placed at our disposal by Dr. N. E. Warburg, Køge.

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[1] Roth, H., and Schuster, Ph., *Angew. Chem.*, **52**, 149 (1939).

Antibodies in Ribonucleoproteins

IN many hypotheses concerning the mechanism of the immune response the nucleic acids have been assigned a role in the production of antibodies, although no clear evidence for their participation is yet available. As regards protein synthesis in general, there is reason for believing that the ribonucleoproteins lie in the pathway of this synthetic process[1]. Evidence also exists that certain enzymes are present in the ribonucleoprotein complex[2,3], and it is possible that they represent products of protein synthesis mediated by ribonucleoprotein. The present communication describes an attempt to ascertain whether antibodies also are present in ribonucleoprotein complexes isolated from the antibody-forming organs, spleen and lymph nodes, of the rabbit.

Evidence that ribonucleoprotein may be involved in antibody production has already been presented by Sterzl and Hrubesova[4] and Hrubesova *et al.*[5].

They have shown that infant rabbits, still unable to synthesize antibodies, acquire this ability when injected with crude 'nucleoproteins' obtained from the spleens of immunized adult rabbits. The 'nucleoproteins' used in this important and suggestive work were, however, very heavily contaminated with extraneous protein. Furthermore, it is difficult to distinguish between the contribution of donor and recipient animal in this type of experiment. We have therefore employed more highly purified ribonucleoprotein preparations and have attempted an *in vitro* demonstration of antibodies.

The experiments were carried out in the following way. Immunized rabbits (together with several non-immunized control animals) were killed when they showed a high level of serum antibodies. The spleens and sometimes the lymph nodes were removed and used for the isolation of ribonucleoprotein. It was generally found that these organs were larger and heavier in the immunized animals than in the controls.

The isolation of ribonucleoprotein was carried out in a cold room, and a buffer containing 0·002 *M* magnesium sulphate and 0·001 *M* *tris*(hydroxymethyl)aminomethane, *p*H 7·1, was used throughout. Microsomes were prepared from homogenized tissues by a low- (10,000*g*, 30 min.) and a high- (105,000*g*, 2 hr.) speed centrifugation. The microsomes were disrupted with 0·75 per cent sodium deoxycholate, and ribonucleoprotein particles were isolated by two further cycles of high- and low-speed centrifugation[6]. The ribonucleic acid content of the purified material was determined as described elsewhere[5]. As weight per cent of ribonucleic acid plus protein it amounted to about 30–40 per cent for the spleen ribonucleoprotein and 45–50 per cent for the lymph node particles. In nearly all cases the yield of ribonucleoprotein per unit weight of organ was higher for the immunized than the control animals.

Portions of the ribonucleoprotein were digested with pancreatic ribonuclease (1–3 μgm./ml.) under dialysis against several changes of 0·15 *M* sodium chloride at 37° for 5–6 hr., followed by an overnight dialysis in the cold. This treatment removed virtually all the ribonucleic acid. Although the bulk of the protein coagulated, a certain proportion remained soluble. After removal of the coagulum by centrifugation, the soluble protein derived from the degraded nucleoprotein was tested for the presence of antibodies. A number of such tests were also made with undegraded ribonucleoproteins.

In most of our experiments the antigen was sheep red blood cells and the tests were made only with spleen ribonucleoprotein. In these cases the animals were immunized with 1 ml., respectively, of a 30 and 15 per cent suspension of red blood cells injected intravenously six days apart. Six days after the second injection the serum antibody titres of these animals and of the controls were determined, the spleens were removed, and ribonucleoprotein was isolated. The serum titre of the immunized animals was generally of the order of 2^{10}. Direct tests for the presence of agglutinins were made by incubating serial dilutions of either the intact ribonucleoprotein or the soluble protein fraction in physiological saline at 37° with 1 per cent suspensions of the antigen.

No trace of agglutinins was detected in the intact ribonucleoprotein, whether from non-immunized or immunized animals. When, however, the soluble protein fraction derived from the degraded ribonucleoprotein of immunized animals was tested, a typical agglutinin reaction was observed, with titres up to 2^4. In contrast, the soluble proteins from the non-immunized controls caused no agglutination. Thus, antibodies appear to have been present in the spleen ribonucleoproteins of the immunized animals. Their immunological reactivity was apparently not expressed until the ribonucleoprotein was degraded, at which time, presumably, the antibody would be liberated from the nucleoprotein complex.

In view of the low agglutinin titres found in the direct tests, the soluble protein fractions were also subjected to the more sensitive Coombs test. Here, 3 per cent suspensions of sheep erythrocytes (in 0·14 *M* sodium chloride buffered at *p*H 7·1 with 10^{-3} *M* *tris*(hydroxymethyl)aminomethane) were incubated with the soluble proteins for 90 min. at 37°, were washed with buffered saline, and were tested against two times serial dilutions of sheep anti-rabbit serum ('Wellcome' sheep precipitating serum). Those erythrocytes which had been incubated with the soluble ribonucleoprotein proteins of immunized animals were completely agglutinated, being shown thereby to be coated with rabbit antibodies. The Coombs reaction gave a titre of 2^9, with a pro-zone of 2–3 dilutions. Again, the non-immunized controls gave negative tests.

In another experiment non-agglutinating antibodies were tested for. Horse serum globulins, mixed with Freund adjuvant, were injected subcutaneously into rabbits. Nine days later, when the serum titre >3125, spleen and lymph node (inguinal, axillar and brachial) ribonucleoproteins were prepared and degraded with ribonuclease. As noted above, the yield of ribonucleoprotein per unit weight of organ was markedly greater for the immunized than for the non-immunized animals (1·09 vs. 0·53 mgm./gm. of lymph node tissue). Direct interfacial ring tests of the soluble protein fractions gave negative results. Tests were then made with Stavitsky's modification[7] of Boyden's method, wherein the antibody agglutinates antigen-coated sheep erythrocytes (tanned at 37°, sensitized with antigen by exposure to horse serum at room temperature, washed and suspended in a hundred times diluted complement-inactivated (56°, 30 min.) non-immune rabbit serum).

The results (Table 1) show that antibodies were present in the soluble protein fraction derived from the lymph node ribonucleoprotein of immunized animals. The absence of detectable antibodies in the spleen ribonucleoprotein is of doubtful significance, owing to the possibility that the subcutaneous immunization had caused the nodes to be activated earlier than the spleen and to have undergone a more intense response at the time of organ extirpation.

Although the experiments described here are still in a preliminary stage, they indicate that the process of antibody response involves a marked increase in the ribonucleoprotein content of the antibody-

Table 1. AGGLUTINATION BY RABBIT RIBONUCLEOPROTEINS OF TANNED ERYTHROCYTES COATED WITH ANTIGEN*

Dilution of antibody solution	×1	×2	×4	×8	×16
Antibody solutions					
Soluble proteins from ribonucleoprotein of:					
(1) Lymph nodes, non-immunized	–	–	–	–	–
(2) " " immunized	+++	++	+	–	–
(3) Spleen, non-immunized	–	–	–	–	–
(4) " immunized	–	–	–	–	–
Controls					
(5) 100 times diluted antiserum	+++	+++	++	+	–
(6) 100 times diluted normal serum	–	–	–	–	–

* Antigen: horse serum globulins.

forming organ, and that the protein moiety of this ribonucleoprotein contains antibodies which are released in an active form upon the destruction of the ribonucleic acid moiety. These observations are consistent with the concept that the antibody molecule forms part of the ribonucleoprotein complex within the antibody-producing cell, and that this complex may, therefore, be the site of its synthesis Similar observations have been made in the case of the release of the enzymes ribonuclease and deoxyribonuclease from a bacterial ribonucleoprotein[2], and it is possible that this is a general phenomenon of protein biosynthesis.

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[1] Littlefield, J. W., Keller, E B., Gross, J , and Zamecnik, P C , *J. Biol. Chem.*, **217**, 111 (1955).

[2] Elson, D., *Biochim. Biophys Acta*, **27**, 216 (1958) , *Biochim. Biophys. Acta* (in the press).

[3] Siekevitz, P., and Palade, G. E , *J. Biophys Biochem Cytol.*, **4**, 309 (1958).

[4] Sterzl, J., and Hrubesova, M., *Folia Biologica*, **2**, 21 (1956)

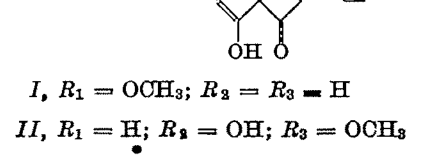
[5] Hrubesova, M , Askonas, B. A., and Humphrey, J. H., *Nature*, **183**, 97 (1959).

[6] Palade, G. E , and Siekevitz, P , *J Biophys. Biochem Cytol* , **2**, 171 (1956).

[7] Stavitsky, A. B., *J. Immunol.*, **72**, 360 (1954).

Flavonoids of the Ponderosa Lemon

THE isolation and structure of citronin, a new flavanone glycoside from the peel of immature Ponderosa lemons (*Citrus limon* Burm. *f. ponderosa* Hort.) was reported some time ago by Yamamoto and Oshima[1]. From chemical and spectroscopic evidence it was concluded that citronin is 2′-methoxy-5,7-dihydroxyflavanone 7-rhamnoglucoside (*I*) and thus belongs to the rather uncommon group of flavonoids having a substituent at position 2′. In attempting to obtain this compound from samples of both mature and immature Ponderosa lemons, under conditions approximating those of Yamamoto and Oshima, it has been our experience that the only glycoside which can be isolated easily in crystalline form is neohesperidin (*II*). This was identified by elementary analyses, mixed melting point determination, infra-red spectrum, preparation of the phenylhydrazone derivative and isolation of the aglycone hesperetin from the hydrolysate. Neohesperidin differs from hesperidin only in the position of attachment of rhamnose to glucose[2]:



$$I,\ R_1 = OCH_3;\ R_2 = R_3 = H$$

$$II,\ R_1 = H;\ R_2 = OH;\ R_3 = OCH_3$$

The isolation of neohesperidin from the Ponderosa lemon is of interest for several reasons. First, as Swingle has pointed out[3], "the nature of the characteristic glucoside present in the tissues of a species of *Citrus* may be of definite taxonomic significance in distinguishing that species from another species to which it may have superficial resemblances". The Ponderosa is generally considered to be in the nature of a hybrid which most nearly resembles the lemon (*Citrus limon*), but it is also regarded as having certain characteristics of the citron and grapefruit[4]. Ordinary lemons have not been shown to contain neohesperidin, but do contain hesperidin, diosmin[5] and eriodictyol glycoside[6] as their principal flavonoids. From qualitative tests, we consider that the Ponderosa lemons used in the present work contained little if any hesperidin or eriodictyol glycoside. Therefore, to judge from the identity of the most abundant and easily detectable flavonoids, the Ponderosa lemon would seem to have little relation to ordinary lemons. It is noteworthy that neohesperidin has been found previously only in the sour orange (*Citrus aurantium*)[7] and trifoliate orange (*Poncirus trifoliata*)[8].

Secondly, although it would be unwarranted to suggest that citronin is identical with neohesperidin (since there appear to be several irreconcilable points of difference), it should be noted that some of the chemical evidence reported by Yamamoto and Oshima is incompatible with their structure *I*. According to these authors, citronin (*a*) gives no ferric colour; (*b*) gives *o*-hydrocoumaric acid methyl ether when cleaved with alkali; and (*c*) after hydrolysis to its aglycone gives 5,7,2′-trimethoxyflavanone on treatment with diazomethane. On the contrary, compound *I* or its aglycone would unquestionably give a ferric colour; would be expected to yield *o*-coumaric acid methyl ether (rather than the hydrogenated derivative); and would undergo methylation at the 5-position very slowly, if at all, by means of diazomethane. In fact, some of the evidence suggests that citronin might conceivably be a dihydrochalcone similar to phloridzin.

Finally, it should be mentioned that botanical variations might account for the difference in the results of Yamamoto and Oshima and ourselves. Dr. W. P. Bitters has pointed out to us that there are two forms of the Ponderosa lemon, one of American origin (used in the present work) and the other of Javanese origin, which are undoubtedly of slightly different genotypes but of very similar phenotypes.

We should like to thank Dr. Bitters and Dr. W. B. Sinclair of the Citrus Experiment Station, University of California, Riverside, for their kindness in providing us with authentic Ponderosa lemons.

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[1] Yamamoto, R., and Oshima, Y., *J. Agr. Chem. Soc. Japan*, **7**, 312 (1931).

[2] Zemplén, G., and Tettamanti, A. K., *Ber.*, **71**, B, 2511 (1938). Horowitz, R. M., and Gentili, B., Abstr. 135th meeting of the American Chemical Society, April, 1959, p. 7 D.

[3] Swingle, W. T., "The Citrus Industry", **1**, ed. Webber, H. J. and Batchelor, L. D., 393 (University of California Press, Berkeley and Los Angeles, 1943).

[4] Webber, H. J., *ibid.*, 605.

[5] Horowitz, R. M., *J. Org. Chem.*, **21**, 1184 (1956).

[6] Horowitz, R. M., *J. Amer. Chem. Soc.*, **79**, 6561 (1957).

[7] Kolle, F., and Gloppe, K., *Pharm. Zentralhalle*, **77**, 421 (1936). Karrer, W., *Helv. Chimica Acta*, **32**, 714 (1949).

[8] Wan, L. K., *J. Pharm. Soc. Japan*, **62**, 466 (1942).

An Elastin Gel

THE present report describes the conversion of insoluble aorta elastin to a gel. Wood[1] recently described the preparation of an elastin gel from a soluble protein derived from cattle ligamentum nuchæ. These observations may have some bearing on the relationship between collagen and elastin[2-5].

Elastin was prepared from cattle aorta by a method described previously[6], in which 1 *N* sodium hydroxide is used for removing collagen. (Some cattle aorta do not yield insoluble elastin under these severe conditions, perhaps owing to their age.) For the preparation of the gel, the elastin was suspended at room temperature in a 0·1 per cent solution of elastase, in acetate buffer at *p*H 4·7 and ionic strength 0·1. (Two different elastase preparations were used: a lyophilized water extract of porcine pancreatin and porcine elastase *PI*[6].) After at least 10 min., the suspension was centrifuged and the residue washed with the buffer. The washed residue was then suspended in 10 vol. of 0·2 *N* acetic acid, from which, on standing, the gel formed.

Gel formation does not occur when (*a*) cattle ligamentum nuchæ is the elastin source; (*b*) cattle aorta elastin is boiled in 0·1 *N* sodium hydroxide; or (*c*) the enzyme treatment is omitted.

The gel melts upon either warming or adding sodium chloride, with subsequent formation of an elastin precipitate. An approximation of the melting point can be made by suspending 1 vol. of packed gel in 5 vol. of aqueous solvent and noting the temperature at which flocculation begins. As the concentration of sodium chloride of the suspension medium increases from 0·01 *M* to 2·0 *M*, the melting and precipitation point first rises and then falls, the maximum occurring between 0·05 and 0·5 *M* sodium chloride. Syneresis has never been observed, even after prolonged storage at 4°C., in the absence of salt.

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[1] Wood, G. C., *Biochem. J.*, **68**, 21P (1958).

[2] Banga, I., *Nature*, **172**, 1099 (1953).

[3] Burton, D., Hall, D. A., Keech, M. K., Reed, R., Saxl, H., Tunbridge, R. E., and Wood, M. J., *Nature*, **176**, 966 (1955).

[4] Burton, A. C., *Physiol. Rev.*, **34**, 619 (1954).

[5] Ramachandran, G. N., and Santhanam, M. S., *Proc. Ind. Acad. Sci.*, **45**, 124 (1957).

[6] Grant, N. H., and Robbins, K. C., *Arch. Biochem. Biophys.*, **66**, 396 (1957).

PHYSIOLOGY

Experimentally Induced Proliferation of the Rudimentary Gonad of an Intact Domestic Fowl

IT is well known that the right female gonad of the domestic fowl ceases to grow about the ninth day of incubation and that it always hypertrophies following sinistral ovariectomy. I have shown[1] that œstrogen could act *in vivo* as the inhibitor of rudiment growth. In view of the anti-œstrogenic effect of 17 α-ethyl-19-nortestosterone ('Nilevar', Searle) upon the chicken's oviduct[2], the effect of this substance upon the rudimentary chicken gonad was investigated. Starting at 60 days of age intact White Leghorn females received 40 subcutaneous injections of 'Nilevar'. The substance was dissolved in a carrier of 5 per cent benzyl alcohol in corn oil and 0·1 c.c. of solution was injected daily. Experimental procedures were analogous to those outlined in the earlier paper[1] and the weights of blotted, fixed (Bouin's fixative) tissue were recorded (Table 1).

Table 1

No. of birds	Treatment	Rudiment weight (mgm.) at 100 days ± standard error of the mean
9	Carrier . ..	5 ± 1
10	1·5 mgm. 'Nilevar'/day	13 ± 1
8	3·0 mgm. 'Nilevar'/day	20 ± 2

Statistical analysis showed the differences due to treatment to be highly significant.

This is the first report of experimentally induced proliferation of the rudimentary intact fowl gonad. Detailed results, including evidence that the proliferation is due to the anti-œstrogenic action of 'Nilevar', will be published elsewhere.

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[1] Kornfeld, W., *Anat. Rec.*, **130**, 619 (1958).

[2] Zarrow, M. X., Peters, L. E., and Caldwell, A. L., jun., *Ann. N.Y. Acad. Sci.*, **71**, 532 (1958).

Effect of Testosterone Propionate and Insulin on Muscle Cholinesterase Activity

VARIOUS hormones influence the cholinesterase activity of tissues[1,2]. The purpose of the present work was to investigate the influence of some anabolic hormones such as testosterone and insulin on muscle cholinesterase activity.

Muscle cholinesterase was studied on male albino rats weighing 250 gm. and kept on a standard stock food. The animals were divided into three groups. One group received 1 mgm. testosterone propionate in 0·2 c.c. sesame oil per animal for ten days, the second group 2 I.U. of crystalline insulin per kgm. body-weight for the same period. The third group was used as control. The animals were autopsied 24 hr. after the last injection, and the cholinesterase activity was determined in diaphragm and gastrocnemius in tissue homogenates by Michel's method[3]. The results (Table 1) are expressed in Δ *p*H/hour per gm. of wet tissue weight.

Table 1. EFFECT OF TESTOSTERONE PROPIONATE AND INSULIN ON RAT'S GASTROCNEMIUS AND DIAPHRAGM CHOLINESTERASE

	No. of animals	Gastrocnemius	Diaphragm
Control	24	0·783 ± 0·068	0·910 ± 0·070
Testosterone propionate (1 mgm. per day)	12	1·410 ± 0·110 ($P < 0{\cdot}01$)	1·065 ± 0·050 ($P < 0{\cdot}05$)
Insulin (2 I.U. per kgm. body-weight per day).	12	1·157 ± 0·048 ($P < 0{\cdot}01$)	1·204 ± 0·064 ($P < 0{\cdot}05$)

The cholinesterase activity is expressed in Δ *p*H/hour per gm. of wet tissue. The determinations were carried out on 0·2 gm. wet tissue.

As is seen in Table 1, testosterone propionate produces a significant increase in cholinesterase activity of gastrocnemius ($P < 0{\cdot}01$) and a rather moderate increase in diaphragm cholinesterase activity

($P < 0.05$). Insulin has virtually the same effect (Table 1). An increase in muscle cholinesterase activity is also observed during embryonic development where marked anabolism occurs[4]. Therefore a relationship is suggested between anabolism and cholinesterase activity in striated muscles. The above results seem to support such a hypothesis since testosterone has a well-known myotropic effect[5] and insulin is also considered as an anabolic agent[6].

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[1] Kruskemper, H. L., and Dirscherl, W., *Bioch. Z.*, **323**, 505 (1953).

[2] Kahlson, G., and Renvall, S., *Acta Physiol. Scand.*, **37**, 159 (1956).

[3] Michel, O., *J. Lab. and Clin. Med.*, 34, 1564 (1949).

[4] Nachmansohn, D., *C.R. Soc. Biol. (Paris)*, **127**, 670 (1938).

[5] Kochakian, C. D., and Tillotson, C., "Hormones and Ageing Process", 3 (New York: Acad. Press, 1956).

[6] Lukens, F. D. W., and McCan, S. M., Int. Symposium on "The Hypophyseal Growth Hormone" (New York: McGraw-Hill, 1955).

Assaying Luteinizing Hormone

THERE is a demand for an easy and accurate method of assaying the luteinizing fraction of gonadotrophic hormone. The present method depends upon the formation of corpora hæmorrhagica in the ovaries of immature female mice after rendering their Graafian follicles mature. The gonadotrophic materials used were pregnant mare serum ('Antex', Leo) as a source of follicle stimulating hormone, and chorionic gonadotrophin ('Physex', Leo) as a source of luteinizing hormone. The material used for assay was dissolved immediately before use in sterile saline and administered as a single dose to avoid deterioration of the hormone by storage even when refrigerated[1].

Table 1. INFLUENCE OF DOSE OF PREGNANT MARE SERUM ON THE CORPUS HÆMORRHAGICUM RESPONSE

Dose of PMS (I.U.)	Dose of HCG (I.U.)	No. of animals	Average No. of corpora hæmorrhagica/mouse*
20	None	15	0·20 ± 0·03
20	10	15	3·66 ± 0·26
40	None	15	0·28 ± 0·07
40	10	15	3·86 ± 0·10
80	None	15	0·30 ± 0·03
80	10	15	3·43 ± 0·36

* ± Standard error.

In order to determine the dose of pregnant mare serum and the time after its administration when the Graafian follicles become fully developed and ready for ovulation, 90 immature female Swiss mice, 3–4 weeks old, were used. They were divided into 3 groups which were injected subcutaneously with a single dose of either 20, 40, or 80 I.U. of pregnant mare serum. The number of animals with opened vaginas were counted daily. On the fourth day there were more than 80 per cent of these. Half the animals of each group were injected subcutaneously with a single dose of 10 I.U. of chorionic gonadotrophin and vaginal smears were taken daily. Forty-eight hours after administration, all smears were either metœstrus or diœstrus. The animals were killed with ether, their ovaries dissected, and the number of corpora hæmorrhagica formed in both ovaries of each animal was recorded.

After establishing the time relations of the assay method, 4 runs of assay were planned. Two assays were run together. In one case the initial dose of pregnant mare serum was 40 I.U. and in the other 20 I.U. After four days the animals were given graded doses of chorionic gonadotrophin as shown in Table 2. The third assay was planned to determine if variations in the follicle-stimulating hormone/luteinizing hormone ratio would influence the slope of the regression line and if it induces a source of error in the assay. 4 days after the injection of 20 I.U. of pregnant mare serum as usual, a total dose of 25 I.U. of different combinations of pregnant mare serum and chorionic gonadotrophin were injected as presented in Table 3. The assay No. 4 was performed to test if blood serum could modify or augment the corpus hæmorrhagicum response to luteinizing hormone as happens in other methods[2]. A comparison was made between the corpus hæmorrhagicum response to 10 I.U. of chorionic gonadotrophin dissolved in saline with that dissolved in ox serum (Table 4).

Table 2. CORPUS HÆMORRHAGICUM RESPONSE TO GRADED DOSES OF CHORIONIC GONADOTROPHIN

Dose of HCG (I.U.)	Average No. of corpora hæmorrhagica 40 I.U. PMS	20 I.U. PMS*
None	0·28 — 0·07	0·33 ± 0·10
1	2·86 — 0·09	3·28 ± 0·33
10	4·86 — 0·48	5·83 ± 0·28
100	8·93 — 0·85	9·83 ± 0·84
1,000	—	9·25 ± 0·98

* ± Standard error. Each group contained 7 mice.

Table 3. INFLUENCE OF FOLLICLE STIMULATING HORMONE/LUTEINIZING HORMONE RATIO ON CORPUS HÆMORRHAGICUM RESPONSE

Dose of PMS (I.U.)	Dose of HCG (I.U.)	Average No. of corpora hæmorrhagica
None	None	0·33 ± 0·05*
5	20	6·00 ± 0·28
10	15	5·00 ± 0·33
15	10	4·50 ± 0·38
20	5	3·80 ± 0·36

* ± Standard error.
Each group contained 7 animals.
All mice were previously treated with 20 I.U. of PMS.

Table 4. INFLUENCE OF ADDITION OF SERUM ON THE CORPUS HÆMORRHAGICUM RESPONSE

Treatment	Average No. of corpora hæmorrhagica*
0·5 ml. ox serum	1·25 ± 0·12
10 I.U. of HCG + 0·5 ml. ox serum	5·00 ± 0·86
10 I.U. of HCG + 0·5 ml. saline	4·75 ± 0·12

* ± Standard error.
Each group contained 8 animals.
The animals were previously treated with 20 I.U. PMS.

The average of the number of corpora hæmorrhagica formed in an animal was plotted against the logarithm of the dose. This dose response curve showed a straight line, up to 100 I.U. of chorionic gonadotrophin. The slopes of the response curves constructed under these conditions were estimated and tests of the hypothesis of equality of the regression lines were performed. The results presented in Tables 1 and 2 show that the initial dose of follicle-stimulating hormone did not modify the corpus hæmorrhagicum response to luteinizing hormone administration. When 20 or 40 I.U. of pregnant mare serum were used, the slopes of the two regression lines were not different ($t = 0.19$). The results presented in Table 3 show that variations in follicle-stimulating hormone/luteinizing hormone ratio of the test material had no influence on the number of corpora hæmorrhagica formed by luteinizing hormone. It appears also that ox serum did not affect the ovarian response to luteinizing hormone. The passage of the ovaries through susa, alcoholic iodine, alcohol, methyl benzoate and finally benzene renders them transparent and corpora hæmorrhagica could be easily counted.

Thanks are due to Mr. Abd-El-Wahab S. Ahmed, of the United States Naval Medical Research Unit No. 3, Cairo, Egypt, for supplying us with the animals used in this investigation.

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[1] Maddock, W. O., and Heller, C. G., *Endocrinol.*, **41**, 177 (1941).

[2] Diczfalusy, E., *Acta Endocrinol.*, **17**, 58 (1954).

Carbon Dioxide Assimilation by *Leishmania donovani*

WHILE studying the glucose metabolism of *Leishmania donovani*, the protozoan organism causing visceral leishmaniasis, it has been found that cell suspensions of this organism in a saline-bicarbonate buffer containing glucose can assimilate a measurable amount of carbon dioxide along with the formation of different acidic end products[1].

L. donovani, strain 81, cultured on Ray's medium[2] at 22–24° C. and recycled every three weeks from the Nicolle-Novy-McNeal medium, was used in this study. After growth for three days on Ray's medium, the organism was scraped from the surface of the agar, washed twice with ice-cold isotonic saline by centrifugation at 3,000 r.p.m. for 10 min. and finally resuspended in isotonic saline.

Standard conditions used for the measurement of carbon dioxide assimilation were as follows: 3·0 ml. of the complete reaction mixture contained 15 μ moles of sodium bicarbonate (*p*H 7·3), 30 μ moles of glucose, 1 ml. of cell suspensions in isotonic saline containing 1·5–2 mgm. protein nitrogen. Incubations were carried out for one hour in conventional Warburg manometric vessels at 37·2° C. in an atmosphere 95 per cent nitrogen and 5 per cent carbon dioxide.

Results from three typical experiments showing the carbon dioxide balances during anaerobic glucose breakdown by cell suspensions of *L. donovani* are presented in Table 1. It will be seen that the amount of carbon dioxide assimilated by the organisms under these conditions varies from 32 μl. to 80 μl. The variation in the amount of carbon dioxide fixation was afterwards found to be due to the different growth stages of the culture used. Table 2 represents figures for the stoichiometric relationship between glucose utilized, acid produced and carbon dioxide fixed in three typical experiments carried out under standard experimental conditions. It will be seen that nearly 0·30–0·36 mole of carbon dioxide is incorporated per mole of glucose consumed. Results using growing culture indicate that the amount of carbon dioxide fixed varies with the age of culture; with two days old growing culture the molar ratio of carbon dioxide fixed per mole of glucose utilized is nearly one. From Table 2 it will also be seen that an average of 1·5 moles of acids are formed per mole of glucose metabolized. Chemical analysis of the different acids that are formed indicated that the major products were non-volatile in nature, and among them succinic acid predominated although malic acid formation has also been noted.

Table 1. CARBON DIOXIDE BALANCES DURING ANAEROBIC BREAKDOWN OF GLUCOSE BY *L. donovani* SUSPENSIONS

No of Exp.	* Total available carbon dioxide (μlitre)	Carbon dioxide liberated due to acid formation	Residual carbon dioxide	Carbon dioxide fixed
	a	*b*	*c*	*a*–(*b* + *c*)
1	262	144	74	44
2	267	150	37	80
3	126	82	12	32

Table 2. CARBON DIOXIDE ASSIMILATION BY *L. donovani* SUSPENSIONS

Expt. No	Glucose consumed (μmole)	Total acid formed (μmole)	Carbon dioxide fixed (μmole)	Carbon dioxide fixed/ Glucose used
1	3·1	4·4	1·1	0·36
2	6·1	8·6	1·9	0·31
3	5 3	7 8	1·6	0 30

When glucose was replaced by glycerol, ribose or pyruvate, no measurable amount of carbon dioxide fixation was noticed. Of the various inhibitors tried, DNP (10^{-4} *M*), α-α′-dipyridyl (10^{-3} *M*) and 8-hydroxyquinoline (10^{-3} *M*) were found to have marked inhibitory effects on the glucose utilization as well as on the carbon dioxide assimilation. It seems that the carbon dioxide assimilation by this protozoal organism is in some way related to the glucose metabolism. The high rate of carbon dioxide assimilation may also be complementary to the very active transaminase system reported to be present in this organism[3].

Detailed studies on the biochemical basis for the carbon dioxide requirement of this protozoal organism are in progress and will be reported elsewhere.

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[1] Chatterjee, A. N., Ray, J. C., and Ghosh, J. J., Symposium on Chemotherapy, C.S.I.R., New Delhi, 168 (Nov., 1958).

[2] Ray, J. C., *Ind. J. Med. Sci.*, **29**, No. 2, 355 (1932).

[3] Chatterjee, A. N., and Ghosh, J. J., *Nature*, **180**, 1425 (1957).

HÆMATOLOGY

Hæmolytic Properties of Immune Chicken Antibody

IN the course of a study of immunological responsiveness of chickens to foreign red blood cells our interest was attracted by the observed discrepancy between high agglutination titres of immune chicken sera and their inability to show lytic properties in the presence of complement. It is well known that different antibody–antigen systems exhibit different abilities to fix guinea pig complement when mammals are used as antibody producers[1]. The failure of immune bird sera to show a significant complement-fixing activity was mentioned by Meyer[2] and by Rice[3] in birds infected with ornithosis virus and *S. pullorum*. It was postulated that avian antibody formed with antigen non-complement-fixing aggregates[4], and that avian antibody lacked some character that was important for combining with complement[5]. This problem, however, was not further explored, and all available data were based on the experiments made with guinea pig complement. It seemed, therefore, reasonable to investigate in a preliminary way whether or not chicken antibody hæmolyses red cells in the presence of complement from different animal species.

Hæmolytic sera were prepared by inoculating intravenously Rhode Island Red pullets with 5 per cent suspension of duck and rabbit erythrocytes (two series of three injections each, followed by re-injection after 20 days). The diluent for all reagents was sodium chloride–veronal buffer *p*H 7·3–7·4 containing

Table 1. HÆMOLYTIC ACTIVITY OF NORMAL ANIMAL AND IMMUNE AVIAN SERA IN THE PRESENCE OF COMPLEMENT FROM VARIOUS SOURCES

Normal animal serum				Immune chicken serum (hæmolysins)	
Species (source of complement for immune hæmolysis)	Complement units in 1 ml.	Hæmolysins for duck red cells	Hæmolysins for rabbit red cells	anti-duck*	anti-rabbit*
Chicken	10·41	–	+1 : 25	–	×
Duck	24 34	–	+1 : 32	–	×
Owl	3·03	–	+1 : 10	–	×
Swan	31·40	–	+1 · 20	–	×
Eagle	32·47	–	+1 : 32	–	×
Wild goose	30 0	–	+1 : 32	–	×
Pelican	34·70	–	+1 : 32	–	×
Ground squirrel	<1·0	–	–	–	–
Guinea pig.	180·50	–	–	–	–
Rat	13 45	–	–	–	–
Rabbit	24·45	–	–	–	–
Dog	55·52	+1 : 25	–	×	–
Cat	6·63	+1 32	+1 : 16	×	×
Horse	<1·0	–	–	–	–
Ox	2 49			–	–
Sheep	<1·0	–	–	–	–
Pig	18·21	+1 : 2	+1 : 2	–	–
Rhesus monkey	66 45	+1 40	+1 40	×	×
Man	32·38	+1 : 32	+1 : 32	×	×
Frog	56·40	+1 : 8	+1 : 32	–	×

–, No hæmolysis; +, hæmolysis; ×, the hæmolytic reaction was not done because of the presence of normal hæmolysins in the quantity of the serum required as complement source.

* In all cases the complement units were present in the supernatants after incubation.

optimal concentrations of calcium and magnesium ions[6]. Sheep, duck and rabbit red cells (3 per cent suspension; *O.D.* 0·453; Beckman spectrophotometer at 541 mμ) were standardized as in Osler *et al.*[7] Pools of fresh sera from different animal species served as the source of complement. The sera were stored at – 65° C. The hæmolytic complement activity of pools was titrated with sheep red cells sensitized with rabbit hæmolysin, and the titre estimated in 50 per cent hæmolytic units. Two and five 50 per cent complement units were used in titration of chicken antisera. The dilutions of antisera were in the range of 1 : 2 to 1 : 3,000. Duck and rabbit erythrocytes were sensitized in chicken anti-duck and anti-rabbit sera, respectively, for 30–90 min. The incubation with two or five complement units lasted 60–90 min. All reactions were carried out at 37° C. Complement hæmolytic units left in supernatants were estimated with sensitized sheep erythrocytes (rabbit hæmolysin). The chicken immune sera were also examined for the presence of agglutinins, and pools of animal sera were investigated for the presence of normal hæmolysins for duck and rabbit erythrocytes.

Agglutination titres of immune chicken sera were 1 : 1,000–1 : 4,000 for rabbit, and 1 : 120–1 : 1,000 for duck red cells. Some animal sera were very poor in complement (ground-squirrel, horse, sheep, ox and owl). It is evident that complement of different animal species was involved in the hæmolysis of sensitized sheep erythrocytes. On the other hand, no hæmolytic activity of chicken anti-duck and anti-rabbit sera could be observed in the presence of complement. It is interesting to note that normal hæmolysins in avian and animal sera exhibit hæmolysis. This observation may suggest that some differences exist between normal hæmolysins and immune hæmolysins in avian sera.

It is difficult to believe that avian species are not able to produce immune hæmolysins. The failure of immune chicken antibody to hæmolyse red blood cells is probably due to its peculiar behaviour in fixing complement. It should, however, be borne in mind that the complement-fixation is a multiple step reaction of very complex nature which may be affected by a variety of conditions, and in which the complement components react disproportionately[8]. Our knowledge about complement reaction is based mainly on the data obtained from the experiments with guinea pig complement and sensitized sheep erythrocytes. But one is confronted with a somewhat different situation when working with avian antibody. The present experiments suggest that immune hæmolysis in which avian antibody is involved differs to some extent from other immune hæmolyses and requires further study which will probably reveal important information about the mechanism of the action of complement.

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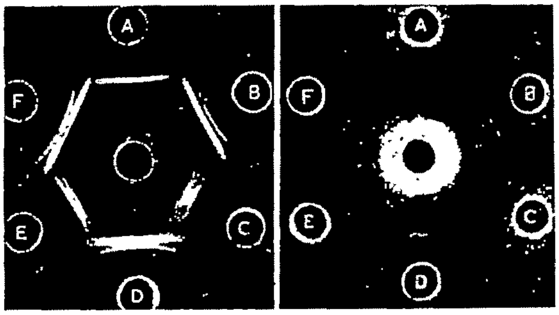
[1] Kabat, E. A., and Mayer, M. M., "Experimental Immunochemistry", 130 (Charles C. Thomas Pub., Springfield, 1948).
[2] Meyer, K. F., in "Diseases of Poultry", 455 (Iowa State College Press, Arwes, 1944).
[3] Rice, C. E., *Canad. J. Comp. Med.*, 11, 236 (1947).
[4] Rice, C. E., *J. Immunol.*, 59, 365 (1948).
[5] Meyer, K. F., and Eddie, B., in "Diagnostic Procedures for Virus and Rickettsial Diseases", 399 (Pub. Office, Amer. Public Health Assoc., New York City, 1956).
[6] Mayer, M. M., and Levine, L., *J. Immunol.*, 72, 511 (1954).
[7] Osler, A. G., Strauss, J. H., and Mayer, M. M., *Amer. J. Syph.*, 36, 140 (1952).
[8] Osler, A. G., *Bact. Rev.*, 22, 246 (1958).

Individual Specific Nature of the Cold Auto-Antibodies of Acquired Hæmolytic Anæmia

DURING the course of an immuno-chemical study of the cold agglutinins present in the sera of patients suffering from acquired hæmolytic anæmia of the cold-antibody type, evidence was obtained which indicated that they were immunologically distinct.

Sera from seven patients were available. Antisera were prepared in rabbits, (1) against the cold antibodies of two of these patients, eluted from erythrocyte stroma, (2) against separated cryoglobulin (acting as a cold antibody) derived from one patient, (3) against γ_1-globulin separated by starch-block electrophoresis from the serum of a patient with acquired hæmolytic anæmia secondary to lymphosarcoma, and (4) against γ_1-globulin separated from a normal serum. The antisera were tested for their reactions by double diffusion in agar gel and immuno-electrophoretic techniques[1].

When the rabbit antisera produced against the pathological cold agglutinins were absorbed with 2·5 mgm. per ml. of γ-globulin (Lister Institute, London) and tested by double diffusion in agar gel against the homologous and heterologous cold-antibody-containing sera and a normal serum, the normal serum produced a single line of precipitate whereas each of the pathological sera produced two lines of precipitate (Fig. 1*a*). The lines formed by the homologous serum showed a reaction of partial identity (cross-reaction) with those formed by the heterologous sera or normal serum, as indicated by spur formation. This suggested the presence of individually specific groupings in the homologous cold-agglutinin protein.



a *b*

Fig. 1. *a*, Photograph of an agar-gel-diffusion plate showing the precipitation pattern of a human serum containing high-titre cold agglutinin (cups *B*, *D* and *F*) and two other human sera also containing high-titre cold agglutinins (cups *C* and *E*) and a normal human serum (cup *A*) The central cup contains rabbit antiserum against the cold agglutinin of the serum used in cups *B*, *D* and *F* and was absorbed with a minimal amount of γ-globulin. The human sera were diluted 1 in 4. *b* Photograph of an agar-gel-diffusion plate showing the precipitation pattern of a human serum containing high-titre cold agglutinin (cups *B*, *D* and *F*) and two other human sera also containing high-titre cold agglutinins (cups *C* and *E*) and a normal human serum (cup *A*). The central cup contains rabbit antiserum against the cold agglutinin of the serum used in cups *B*, *D* and *F* and was absorbed with twice its volume of normal serum (central cup). The human sera were diluted 1 in 4

When the antisera were further absorbed with an excess of pooled normal human serum or with the γ_2- and γ_1-globulin fractions separated electrophoretically from normal human serum, they reacted with homologous serum only (Fig. 1*b*). No reaction was discernible with the heterologous pathological sera or normal human serum even when the gel-diffusion plates had been kept at 37° C. for 3 months. Exactly similar reactions were obtained when the cold agglutinins eluted from erythrocyte stroma were used as test antigens in place of the pathological sera.

Further experiments showed that the formation of precipitin lines by the homologous serum only could not be explained by non-specific precipitation due to the presence of heat-labile components of complement, by incomplete absorption of the antisera or by the presence of different globulin groups[2] in the serum used for absorption and the patient's serum. It is of interest to note that when the rabbit antiserum produced against a normal γ_1-globulin was similarly absorbed, it failed to react with any normal or pathological serum, indicating complete neutralization of the antibodies contained in it.

Although specific globulins have been identified by their Gm groups[2] and specific differences reported between the macroglobulins of Waldenstrom's macroglobulinæmia[3], globulins with antibody properties are not usually considered as having individually specific characteristics. The immunological differences observed may have a counterpart in the subtle differences in serological properties already observed by Dacie and de Gruchy[4] in the antibodies of acquired hæmolytic anæmia. The presence of antigenic groupings specific to the cold antibodies of each individual is in accord with the hypothesis that the formation of cold antibodies represents the synthesis of abnormal proteins rather than an immune response on the part of normal antibody-forming tissues. The observation could perhaps be taken to favour the concept of somatic mutation occurring in the antibody-forming tissues of the patient.

I am indebted to Dr. J. H. Humphrey, of the National Institute for Medical Research, Mill Hill, London, for advice and helpful criticisms and for the supply of γ-globulin preparations used in this investigation.

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[1] Mehrotra, T. N. (in preparation).

[2] Grubb, R., and Laurell, A. B., *Acta path. microbiol. Scand.*, 39, 390 (1956).

[3] Korngold, L, and van Leeuwen, G., *J. Exp. Med.*, 106, 467 (1957).

[4] Dacie, J. V., and de Gruchy, G. C., *J. Clin. Path.*, 4, 253 (1951).

Distribution of Gm^a and Gm-like among Javanese, Djuka Negroes, and Oyana and Carib Indians

Grubb and Laurell[1] and others have demonstrated that some human sera contain a genetically determined factor capable of inhibiting the ability of serum from selected rheumatoid arthritic patients (RA serum) to agglutinate Rh(+) cells coated with selected incomplete anti-D sera. They called this factor Gm^a and showed that its presence is due to a dominant gene. Thus far various investigators have tested 2,635 Europeans and found that 56·96 per cent were Gm(a+)[2]. Grubb and Laurell[1] found 95 per cent of 74 Eskimos to be Gm(a+), and Steinberg and Giles[3] found 98 per cent of 98 American Negroes to be Gm(a+).

Steinberg and Giles[3] found an RA–anti-D system which tested for a factor which is not Gm^a, Gm^x[3], or Gm^b (ref. 4 and Harboe, M., personal communication). Because its genetic relation to these factors is unknown, the new factor is tentatively called Gm-like. This factor was present in 27·5 per cent of 403 American Negroes and absent in all of 250 Whites. Its presence is determined by a dominant allele.

We have now tested serum from 20 Javanese, 35 Djuka Negroes, 15 Oyana Indians, and 19 Carib Indians for Gm^a and Gm-like. The method of testing was that described by Grubb and Laurell[1]. The samples were collected by Dr. Vincent Butler in Surinam (Netherlands Guiana). The Djuka Negroes are descendants of slaves who escaped into the jungle shortly after importation into Dutch Guiana. The Oyana Indians are an isolate of jungle Indians, living along a river in the interior of Surinam. The Carib Indians are coastal. So far as we know, these people have not intermarried with other ethnic groups.

All 89 samples were positive for Gm^a. The tests for Gm-like were done blind in the sense that all sera were numbered with no indication of the ethnic origin of their donors. The results of the tests for Gm-like are presented in Table 1.

The samples are small; hence, it cannot be established that the difference in frequency of Gm-like(+) among Javanese and Djuka Negroes is significant ($P > 0{\cdot}3$), if indeed the difference is

Table 1. Results of Tests for Gm-like on Sera from Javanese, Djuka Negroes, Oyana Indians and Carib Indians

		Gm-like			
		+		–	
	Total	No.	Percentage	No.	Percentage
Javanese	20	10	50	10	50
Djuka Negroes	35	22	63	13	37
Oyana Indians	15	0	—	15	100
Carib Indians	19	0	—	19	100

significant. There is little doubt, however, that the frequency of Gm-like(+) among the Indians is less than that among the other two groups [P (Javanese–Oyana) $\sim 0\cdot002$, P (Djuka Negroes–Oyana) $< 0\cdot0001$]. Similarly, there is little doubt that the frequency of Gm-like(+) among the Indians is less than that among American Negroes [$\chi^2_{(1)}$ (corrected) $= 4\cdot33$, $P < 0\cdot05$ for Oyana *versus* American Negroes] or that the frequency of Gm-like(+) among the Djuka Negroes is greater than that among the American Negro ($\chi^2_1 = 19\cdot79$, $P < 0\ 001$).

The data are too few to permit a discussion of the ethnic and racial distribution of these genes (*Gm^a^* and *Gm-like*) beyond stating that the frequency of *Gm*^{a} among the coloured peoples seems to be 100 per cent and that *Gm-like* is essentially absent in Whites and possibly in South American Indians and occurs among Javanese and Negroes. Further data are required. The wide variation in frequency of *Gm-like* among various ethnic groups indicates that it may be a useful tool for anthropologists and others interested in population problems.

We are deeply indebted to Dr. Butler for making these sera available to us and to the hospital and laboratory of the Aluminium Co. of America for making their facilities in Surinam available for the collection of the blood specimens used in this study.

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[2] Reviewed in Steinberg, A. G., and Giles, Brenda Dawn, *Amer. J Human Genet.* (in the press).

[3] Harboe, M., and Lundevall, J., *Acta Path. Microb. Scand.*, 45, 357 (1959).

[4] Harboe, M., *Nature*, 183, 1468 (1959).

Absence of the Diego Blood-Group Antigen in the Lapps

BLOOD-GROUP studies in the Lapps have revealed rather unusual distributions within several blood-group systems[1,2]. The Lapps seemed to have a curious mixture in their blood groups of European features on one hand, and Asiatic ones on the other.

The discovery of the Diego blood-group antigen has given a valuable tool in anthropological investigations[3]. As the Diego antigen is confined to Mongoloid populations, and in view of the possibility of the Lapps being related to these populations, it seemed to be of importance anthropologically to determine whether this factor was present in the Lapps.

We have tested a total of 433 blood samples from Norwegian Lapps for the Diego factor. The anti-Di^a serum and the Di(a+) control cells used in the study were generously placed at our disposal by Dr. Miguel Layrisse, of Caracas, Venezuela. The serum reacted strongly by the indirect Coombs's technique against the Di(a+) cells, whereas all the 433 Lapp blood samples gave negative results.

From Sweden, further results on the blood groups of the Swedish Lapps have just been published[4]. These investigations also include Diego grouping on 220 blood samples, all of them giving negative reactions. Pooling the Swedish and Norwegian data, we find that a total of 653 Lapps have been examined for the Di^a factor. The absence of the factor in a sample of this size seems to exclude any close relationship to the Mongoloid populations carrying this antigen. In this connexion we should, however, like to point out that the Diego antigen was also absent in a population which is usually regarded as Mongoloid, namely, Eskimos from the eastern Canadian Arctic[5,6].

The Norwegian Lapp blood samples were also grouped in the A_1A_2BO, MNSs, P, Rh(C,c,C^w,D,E,e,V), Kell, Lewis(Le^a,Le^b), Duffy(Fy^a), Kidd(Jk^a,Jk^b), and Wright blood-group systems. A report of these investigations will be given elsewhere.

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[1] Allison, A. C., Hartmann, O., Brendemoen, O. J., and Mourant, A. E., *Acta Path. Microbiol. Scand.*, 31, 334 (1952).

[2] Allison, A. C., Broman, B., Mourant, A. E., and Ryttinger, L., *J. Roy. Anthrop. Inst.*, 86/1, 87 (1956).

[3] Layrisse, M., *Amer. J. Phys. Anthrop.*, 16, 173 (1958).

[4] Beckman, L., Broman, B., Jonsson, B., and Mellbin, T., *Acta Genet.*, 9, 1 (1959).

[5] Lewis, M., Chown, B., and Kaita, H., *Nature*, 178, 1125 (1956).

[6] Chown, B., Lewis, M., and Kaita, H., *Nature*, 181, 268 (1958).

Haptoglobin in Monkeys

WE have examined the haptoglobin types of a series of blood samples from 27 monkeys (23 *Macaca mulatta*, 1 *Macaca irus* and 3 *Cebus nigrivittatus*). The *Macaca mulatta* and *Macaca irus*, indigenous to India and Java, were purchased from the United States (Trefflich's Bird and Animal Co. Inc.). Since the rhesus monkey species infrequently reproduces itself in captivity, it can be assumed, even though no genetical results were obtained, that these monkeys are probably unrelated. The few *Cebus* samples examined came from the Higuerote (Miranda State) and Tamanaco (Guárico State) regions, in Venezuela.

The method used for the study of haptoglobins and their differentiation is based mainly on the methods of Smithies[1] and Allison and ap Rees[2] and has already been outlined[3]. Thanks to these methods there exists no doubt as to the classification of haptoglobins.

All the monkeys examined, both from Venezuela and from India and Java, were found to have type 1–1 haptoglobin, probable genotype $H_p{}^1/H_p{}^1$.

The number of monkeys used in our studies is relatively small for definite conclusions to be reached. Should future investigation confirm that monkeys have only one type of haptoglobin, then this finding could be phylogenetically significant, since both monkeys and man belong to the same primate order. In all probability there only existed in the past what is now called type 1–1 haptoglobin, from which the other types known at present are derived by mutation. It would be interesting to know, in this connexion, whether any human populations exist the haptoglobin of which belongs to type 1–1 only. If not, one would be inclined to think that this change probably occurred during or after the evolution of the *Homo sapiens* from non-human and early human types. It is not yet known whether such a mutation has or has not actually favoured this evolution.

Several authors[4-6] have dealt with the much discussed problem of whether the polymorphism of present man is either a recent acquisition or has been directly inherited from his non-human ancestors. Studies made on several species of monkeys in relation to their blood groups as well as to their ability to taste phenylthiocarbamide[4,7] have revealed that the genetical polymorphism of man, in this connexion, is a property common to all primates. If our findings are confirmed, we may be facing a gene (H_p) specifically showing a difference between man and other primates.

We wish to thank Dr. Otto Núñez-Montiel and Dr. Frieda Herold, of the Virus Laboratory of this Institute, for the collection of the blood specimens, and Mr. Alfredo Pansini, of the translation and publication department, for the translation of the Spanish text.

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[1] Smithies, O., *Biochem. J.*, 61, 629 (1955).

[2] Allison, A. C., and ap Rees, W., *Brit. Med. J.*, ii, 1137 (1957).

[3] Rodríguez, M. L. G. de, and Arends, T., *Nature*, 181, 824 (1958).

[4] Stern, Curt, "Principles of Human Genetics" (Freeman and Co., San Francisco, 1949).

[5] Ford, E. B., "Genetics for Medical Students", fourth edit. (Methuen and Co., London, 1956).

[6] Allison, A. C., Cold Spring Harb. Symp. Quant. Biol., 20, 239 (1955).

[7] Fisher, R. A., Ford, E. B., and Huxley, J. S., *Nature*, 144, 750 (1939).

A Survey of Abnormal Hæmoglobins in Portuguese Guinea

IN 1957–58 we conducted a survey of abnormal hæmaglobins among the indigenous population of Portuguese Guinea, during which 9,224 blood samples were collected.

In each case, erythrocytes were tested for sickling, and their hæmoglobin was examined by paper electrophoresis of pH 8·6. When fast-moving hæmoglobins were detected these were submitted also to paper electrophoresis at pH 6·5 and compared with standard samples of known hæmoglobin variants. In the case of hæmoglobin *N*, the first sample we collected was kindly classified by Dr. H. Lehmann in London; after this, further instances of hæmoglobin *N* were classified by ourselves by comparison with this first sample. For identification of hæmoglobin *K*, a sample of *K* obtained during a previous survey at St. Thomas and Prince Island, and one of hæmoglobin *J* supplied by Dr. Lehmann were used. The results of the survey are given in Table 1.

The abnormal hæmoblogins found were *S*, *C*, *K* and *N*.

Not infrequently we found a blood to give a positive sickling test, but not to show the presence of hæmoglobin *S* on electrophoresis. In every case, blood was again withdrawn and submitted to the same tests. In every case, hæmoglobin *S* proved once more to be absent on paper electrophoresis, and the sickling test was negative on the second occasion. We consider that sickling-rates among populations based solely on the sickling test are subject to error, due to false sickling.

The frequency of hæmoglobin *S* varies greatly among the different tribes. As a rule we can say that it is much more common among the Moslem tribes than among the Fetichists.

Hæmoglobin *C* was seen only among the Moslem tribes, which are those with closer relationships with the neighbouring countries.

Hæmoglobin *K* was first described by Cabannes and Buhr[1] among the Algerian Kabyles. Later, Robinson *et al.*[2] found it twice in Liberia, calling it hæmoglobin Liberian II. It has been further found by Ager and Lehmann[3] in an East Indian family, and by Edington and Lehmann among 3 Dagombas of the Northern Ghana, by Cabannes and Sansarricq[1] among 2 Senefous of the High Volta, by Cabannes and Gavanou[1] in one Touareg from the Hoggar, and by Trincão *et al.*[4] in 2 individuals from St. Thomas Island. In Portuguese Guinea we found 8 instances of this rare hæmoglobin; 6 among the Moslem tribes, and 2 among the Fetichists.

Table 1

Tribes	Total No. of observations	Hb. *AS*		Hb. *AC*		Hb. *AK*		Hb. *AN*	
		No of cases	Per cent	No. of cases	Per cent	No. of cases	Per cent	No. of cases	Per cent
Bagas* †	8					1			
Balotes	342	1	0·2						
Balantes	776	7	0·9			1	0 12	1	0·12
Banhuns	63	10	15·8						
Beafadas†	797	103	12·9	1	0 13	1	0 13	1	0 13
Bijagos	632	2	0·47			1	0 15		
Cassangas†	144	34	23·6						
Felupes	641	36	5·5						
Fulas	1,558	248	15·9	6	0·4	2	0·13	4	0 25
Jacancas* †	43	4	9 3						
Jalofos* †	1								
Landoman* †	3								
Laibe* †	3								
Mancanha	534	1	0·2			2	0 4	1	0·2
Mandinga†	895	115	12·8	10	1 12	1	0 1		
Manjaco	758	16	2·1			1	0 13	3	0·4
Miefora* †	1								
Nalus†	100	6	6						
Nar* †	2								
Pajadinca†	300	48	16			1	0·33	2	0 66
Papel	1,244	16	1·3						
Saracole	268	44	16 4	4	1·5				
Sosso†	94	3	3 1					2	2 1
Surua* †	10	2							
Tilbonca* †	8	3							
Timene* †	4								

* Tribes living in neighbouring territories and encountered by chance in Portuguese Guinea.
† Moslem tribes.

Hæmoglobin *N* seems to be very rare. It was first seen in 8 members of the Mande tribe of Central Liberia by Robinson *et al.*[2], who called it Liberian I. Ager and Lehmann mention two further instances, one from Ghana and one from Nigeria. Further instances have been reported from Portuguese Guinea. It seems interesting to point out that recent surveys of abnormal hæmoglobins among some 2,000 Nigerians failed to reveal any further example of this rare hæmoglobin variant (personal communication, Dr. H. Lehmann).

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[1] Cabannes, R., and Buhr, J. L., *Sang*, **29**, 201 (1958).
[2] Robinson, A. R., Zuelzer, W. W., Neel, J. V., Livingstone, F. B., and Miller, M. J., *Blood*, **11**, 902 (1956).
[3] Ager, J. M., and Lehmann, H., *Brit. Med. J.*, i, 1449 (1957).
[4] Trincão, C., Almeida Franco, L. T., Coutinho Gomes, M. P. N., and Cordeiro Ferreira, N., *Sangre*, **3**, 51 (1958); Trincão, C., Almeida Franco, L. T., and Nogueira, A. R., *Nature*, **183**, 193 (1959).

PATHOLOGY

Chorioallantoic Membrane Lesions produced by Inoculation of Adult Fowl Leucocytes

INTEREST in the reaction of the chick embryo to inoculation with adult homologous tissues, noted in 1916 by Murphy[1], has been renewed by the investigations of Simonsen[2] and Cock and Simonsen[3], which indicate that the alterations in the embryo, including splenomegaly, hepatomegaly, and hæmolytic anæmia, result from an immunological attack by the adult cells on the embryonic tissues. This communication reports that cell suspensions prepared from adult fowl blood, spleen or thymus, when placed on the chorioallantoic membrane, produce characteristic membrane lesions in addition to splenomegaly and hepatomegaly. The chorioallantoic lesions closely resemble the splenic nodules observed by Isacson[4] which developed in 11–15 day chick embryos following intravenous injection of adult homologous leucocytes or spleen cells. Isacson suggested that the splenic nodules represented foci of adult donor cells which were stimulated to proliferate by contact with foreign embryonic antigens, and which were afterwards walled off and destroyed by the maturing embryo.

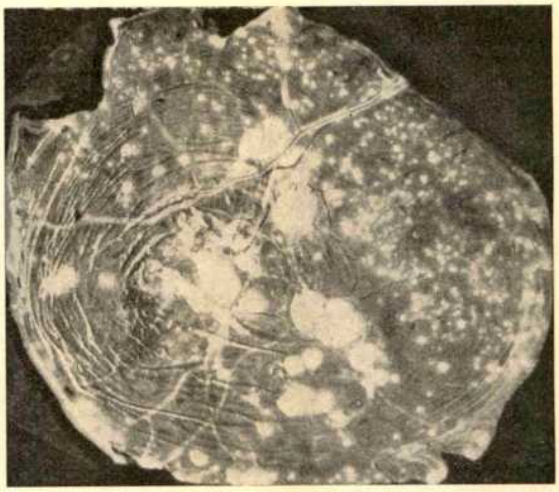

Fig. 1. Chorioallantoic membrane from a chick embryo inoculated at 12 days with a suspension of adult fowl leucocytes and harvested after 6 days incubation at 38° C. Typical large nodules are shown with many small foci which may be of secondary origin.

The chorioallantoic lesions produced by inoculation of adult homologous cell suspensions on to the membrane of 10–14 day old embryos required 4–5 days to develop to the stage illustrated in Fig. 1. At this mature stage they appeared as raised yellow or white nodules, 2–4 mm. in diameter, showing a semi-translucent peripheral zone, and a firm, opaque central core which could be readily shelled out from the surrounding tissue. Multiple tiny opaque areas were sometimes present as well, but they were a less constant feature. Microscopically the inoculated membranes showed a complex variety of changes; only those which appear to be concerned directly with the characteristic nodular lesions will be considered here. The sequence of development of the nodular lesions was similar to that of the splenic nodules described by Isacson[4]. Within 48 hr. after inoculation with adult cells, rounded collections of multiplying mononuclear or spindle-shaped cells were observed in the mesodermal layer of the membrane. By three days, many fibroblasts, mononuclear cells and granulocytes were found surrounding the cell foci. Somewhat later the foci were invaded by granulocytes and underwent necrosis. Infiltration of the adjacent membrane tissue with granulocytes was a frequent and striking feature in the late stages. The alterations in the chick embryo were similar to those which occurred subsequent to intravenous injection of adult homologous blood and spleen cells[2,4]. Any given cell concentration, however, evoked more marked changes in the embryo when given intravenously than when inoculated on to the chorioallantoic membrane. Following either route of inoculation, characteristic nodules developed in the spleen and less frequently in other sites, including the thymus, proventriculus, bone, muscle and subcutaneous tissue. Widespread granulopoiesis occurred, particularly in the periportal areas of the enlarged liver.

The production of the chorioallantoic and associated embryonic lesions by adult fowl cells was a highly reproducible phenomenon. The blood of all the eight adult fowl donors tested, including four in-bred White Leghorn, one New Hampshire Red and three White Leghorn-Australorp hybrid fowls, was effective. As with other manifestations of the donor versus host reaction in avian embryos[2,5], the leucocyte fraction of the blood was found to be responsible for this phenomenon; red blood cells and serum were incapable of producing characteristic nodular membrane lesions. It was also observed that the ability of the leucocytes to induce the lesions was abolished by freezing and thawing, and that white blood cells and spleen cell suspensions from embryonic donors were ineffective. Adult fowl spleen and thymus cell suspensions were capable of inducing characteristic chorioallantoic lesions, although thymic cells appeared less effective than spleen cells and leucocytes.

The number of nodular chorioallantoic lesions which developed depended upon the number of cells inoculated on to the membrane. An inoculum containing 10^4–10^5 peripheral blood leucocytes was required to produce a single nodule. When graded concentrations of cell inocula were given, an approximately proportional relationship was observed between the number of cells introduced and the number of nodular lesions produced. Several experiments have been performed with highly in-bred strains of White Leghorn fowls and embryos. Embryos of the in-bred line responded with approximately the same number of membrane lesions as hybrid White Leghorn-Australorp embryos when inoculated with leucocytes from an unrelated donor fowl. However, when cells from an in-bred donor were placed on the chorioallantoic membranes of isologous embryos, the number of lesions which occurred was far less than the number produced by the same cell suspension on unrelated hybrid embryos.

The investigations made to date indicate that the nodular chorioallantoic membrane lesions result from an immunological reaction between the adult cells and embryonic recipient, presumably of the 'donor versus host' type. The data suggest that the production of lesions requires a significant antigenic disparity between donor and host. In regard to the number of nodules produced, it is a reasonable assumption that each lesion arises as a result of the effective implantation of a single donor cell, but further work will be needed to establish what factors are responsible for the ratio of 10^4–10^5 : 1 between cells inoculated and nodules observed. It may be that a large fraction of the leucocytes are potentially capable of nodule formation but that essentially accidental factors allow only a minute proportion actually to initiate production. At the other extreme, it may be that a very small proportion of cells predetermined to react with the effective isoantigens are responsible for the lesions.

Irrespective of the way the nodules are initiated, it seems legitimate to regard them as arising by proliferation of immunologically competent donor cells in response to antigenic stimulation by components of the embryo. If this view is correct, the phenomenon of chorioallantoic nodule formation should provide a useful approach to certain of the problems of current immunological theory, and may be particularly relevant to the clonal selection hypothesis of Burnet[6].

I wish to express my sincere gratitude to Sir Macfarlane Burnet for his stimulating interest and advice in this work.

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[1] Murphy, J. B., *J. Exp. Med.*, **24**, 1 (1916).
[2] Simonsen, M., *Acta Path. Microbiol. Scand.*, **40**, 480 (1957).
[3] Cock, A. G., and Simonsen, M., *Immunol.*, **2**, 103 (1958).
[4] Isacson, E. P. (to be published).
[5] Terasaki, P. I., Cannon, J. A., and Longmire, W. P., jun., *Proc. Soc. Exp. Biol. Med.*, **100**, 639 (1959).
[6] Burnet, F. M., "The Clonal Selection Theory of Acquired Immunity" (Cambridge University Press and Vanderbilt University Press, 1959).

Influenza A′ Infection of Cultured Cells of Whole Mouse Embryos

CELLS of whole mouse embryo grown in culture have been used for work with polyoma virus[1,2], and influenza viruses have been grown in cultures of other mammalian tissues such as monkey kidney[3,4], human embryo kidney and lung[5] and bovine embryo kidney[6].

For a recent study of the 'eclipse' phase in the multiplication of influenza *A′* virus, the virus was grown in cells of mouse embryo lung[7]. A greater quantity of tissue can be obtained for culture more easily by using whole mouse embryos, and as is described below, such cultures support the growth of this virus.

Whole mouse embryos taken from stock albino mothers bought from a dealer were weighed, chopped up and treated with trypsin. The cells so released were suspended in a medium consisting of inactivated normal rabbit serum, 1 part, 2·5 per cent lactalbumin hydrolysate, 1 part, and Hanks's balanced salt solution, 8 parts, together with sufficient penicillin, streptomycin and mycostatin to control possible contaminants. Such a cell suspension, containing 5–10 × 10^5 cells per ml., was put into insulin vials in 2 ml. lots and produced monolayer cultures suitable for infection with virus after incubation for 18 hr. at 36·5° C.

Infection was achieved by removing the medium from the cells and, after washing them with balanced salt solution, replacing it with the same volume of a 1 : 50 dilution in 5 per cent normal rabbit serum in balanced salt solution of homogenized mouse lung suspension, representing a later passage of the strain of influenza *A′* used in the earlier work[7]. The cells were held at 36·5° C. for 45 min.; then the infecting suspension was removed and kept for infectivity titration in hens' eggs. The cells were washed in 3 changes of balanced salt solution and their original medium restored. This medium was sampled for infectivity titration 2 hr. and 24 hr. after the first contact of the cells with the infecting suspension.

Table 1. GROWTH OF INFLUENZA *A′* VIRUS IN CULTURED CELLS OF WHOLE MOUSE EMBRYOS, SHOWING EMBRYO WEIGHTS

Experiment	Embryos, Mean wt. (gm.)	50 per cent Egg Infective Dose (ml.)		
		Infecting suspension	Culture medium 2 hr.	Culture medium 24 hr.
1	0·08	$10^{-6.2}$	$10^{-1.7}$	$10^{-3.5}$
2	0·11	$10^{-4.7}$	$10^{-2.5}$	$10^{-3.0}$
3	0·31	$10^{-4.6}$	$10^{-2.3}$	$10^{-4.6}$
4	not recorded	$10^{-4.8}$	$10^{-2.8}$	$10^{-5.6}$

As can be seen from Table 1 the system supported multiplication of the virus, the increase in infectivity for eggs in 22 hr. ranging from 0·5 to 2·8 $\log_{10}$ units. This increase may be compared with the finding that medium removed from infected cells at 2 hr., when its mean 50 per cent egg infective dose in three further experiments was $10^{-3.1}$ ml., was rendered completely non-infective by 22 hr. incubation at 36·5° C. in the absence of cells.

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[1] Eddy, B. E., Stewart, S. E., and Berkeley, W., *Proc. Soc. Exp. Biol. Med.*, **98**, 848 (1958).
[2] Rowe, W. P., Hartley, J. W., Estes, J. D., and Huebner, R. J., *J. Exp. Med.*, **109**, 379 (1959).
[3] Takemoto, K. K., Lynt, R. K., Rowe, W. P., Huebner, R. J., Bell, J. A., Mellin, G. W., and Davies, D. J., *Proc. Soc. Exp. Biol. Med.*, **89**, 308 (1955).
[4] Mogabgab, W. J., Green, I. J., Dierkhising, O. C., and Phillips, I. A., *Proc. Soc. Exp. Biol. Med.*, **89**, 654 (1955).
[5] Mogabgab, W. J., Simpson, G. I., Green, I. J., *J. Immunol.*, **76**, 314 (1956).
[6] Green, I. J., Lieberman, and M., Mogabgab, W. J., *J. Immunol.*, **78**, 233 (1957).
[7] Gostling, J. V. T., *Brit. J. Exp. Path.*, **39**, 679 (1958).

Action of Antibodies and Plasmin on Ehrlich Ascites Tumour Cells

THE mechanism of cell destruction in allergic inflammation is not well understood. Two explanations for production of cell damage have, however, been proposed: (*a*) that the antigen-antibody reaction causes, in some non-specific manner, permeability changes in cells which result in the release of active mediators of inflammation (histamine, etc.) and that the latter are directly involved in the allergic process[1]; (*b*) that the antigen-antibody reaction activates proteolytic enzymes (plasmin, proteases) that injure the cells[2-4].

The second hypothesis has been supported by some experimental evidence; but the exact mechanism of cell destruction by proteolytic enzymes remains to be elucidated. Studies on the effect of cytotoxic antibodies against Ehrlich ascites tumour cells have revealed that the cytopathogenic changes induced in tumour cells by the action of specific antibodies were very similar to those obtained by the action of streptococcal hæmolysins[5]. It was also found that tumour cells treated with streptococcal hæmolysins were completely disintegrated by the presence of small amounts of proteolytic enzymes[5]. Therefore the action of plasmin was tried on tumour cells affected by specific antibodies, so as to obtain some insight into the role of blood proteolytic enzymes in this process. Rabbits were immunized with homogenates of Ehrlich tumour cells according to the method of Salk and Laurent[6]. Four weeks following the first injection, the sera of these rabbits up to a titre of 1 : 20 caused a distinct cytopathogenic effect on tumour cells. The action of antiserum was found to be dependent on complement.

In a typical experiment 0·1 ml. of washed ascites cells (10^8/ml.) were incubated at 37° C. with 0·1 ml. of immune serum and with 0·8 ml. of $M/15$ phosphate saline buffer *p*H 7·4. Five minutes following addition of antiserum, agglutination of the tumour cells occurred; while 10 min. later first signs of cellular damage could already be seen. The cells appeared swollen, the nucleus became visible and was surrounded by a clear cytoplasmic space with granules close to the nucleus. Thirty minutes following the addition of antiserum, the majority of the tumour cells appeared dead, as established by staining with trypan blue. Sixty minutes following the addition of antiserum the total number of the tumour cells was reduced by 25 per cent (Table 1). The remaining cells, though swollen, retained their cytoplasmic membrane, and the nucleus appeared undamaged. Only a very small amount of cell debris could be seen. These results are consistent with the findings of Flax[7] and Goldberg and Green[8], who tested the action of antiserum against Ehrlich and Krebs ascites tumours respectively. The action of human and rabbit plasmin, activated by streptokinase SK–100 units/ml. (Varidase-Lederle), was tested on tumour cells affected by antiserum. Rabbit and human plasminogen were prepared according to the method of Ratnoff[9]. In addition, antiserum was also used as a source of plasminogen. Plasmin was assayed according to the method described by Remmert and Cohen[10]. Plasmin (1–50 units/ml.) was added to Ehrlich tumour cells which had been pre-incubated for 15 min. at 37° C. with antiserum, and which showed distinct swelling. As soon as 10 min. after addition of plasmin, the swollen cells began to disintegrate, and 5 min. later large accumulations of cell debris mingled with ghosts of swollen cells could be seen (Fig. 1). Incubation with increasing concentrations of plasmin caused a proportional increase in the percentage disintegration of the tumour cells (Table 1). Under the same experimental conditions neither plasmin alone (250 units/ml.) nor plasmin incubated with heat-inactivated antiserum (56° C. for 30 min.) or streptodornase (1,000 units/ml. present in 'Varidase' preparations) had any effect on the morphology or total number of cells (Table 1). The leucocytes which were present in the tumour suspensions incubated with antiserum and plasmin appeared undamaged (Fig. 1). If on the other hand plasmin was added to tumour cells either simultaneously or 5 min. after addition of antiserum (before any apparent damage to cells occurred) only few cells appeared swollen and no disintegration of tumour cells was observed. These results are consistent with the findings of various investigators[11] that plasmin destroys complement. This assumption seems plausible since the action of cytotoxic antibodies against Ehrlich tumour cells have been found to be dependent on complement[7]. Thus, to obtain rapid disintegration of cells by the combined action of antiserum and plasmin the antibody-complement complex must first damage the cell membrane. This leads to altered permeability of the cell membrane which may allow plasmin either to penetrate into the cells or to attack cell wall substrates which have been unmasked by the action of antiserum. This would result in the depolymerization of cell components leading to cell disintegration.

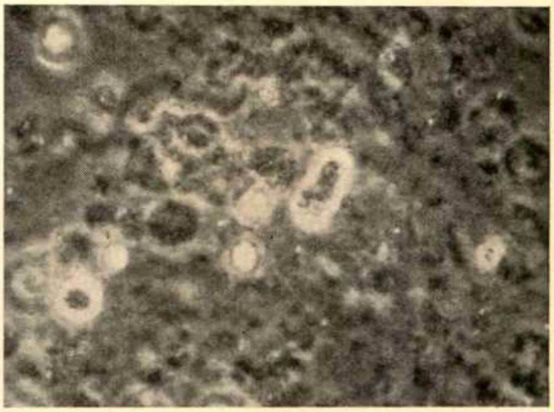

Fig. 1. Ehrlich ascites tumour cells incubated for 15 min. at 37° C. with antiserum and plasmin (50 U./ml.). Plasmin was added to tumour cells after they were in contact for 15 min. with antiserum. Note the massive destruction, accumulation of cell debris, few tumour cell ghosts and several undamaged leucocytes. (× 450)

Table 1. ACTION OF ANTISERUM AND HUMAN PLASMIN ON EHRLICH TUMOUR CELLS

Reaction mixture	Cells remaining* (per cent)
Tumour cells in buffer[a]	100
,, ,, + 10 per cent antiserum[b]	75
,, ,, + 10 per cent inactivated antiserum[c]	100
,, ,, + Plasmin (250 U/ml.)	100
,, ,, + 10 per cent antiserum + plasmin (1 U./ml.)[d]	54
,, ,, + 10 per cent antiserum + plasmin (10 U./ml.)	10
,, ,, + 10 per cent antiserum + plasmin (20 U./ml.)	1
,, ,, + 10 per cent antiserum + plasmin (50 U./ml.)	0·01
,, ,, + 10 per cent antiserum (inactivated) + plasmin (50 U./ml.)	100
,, ,, + 10 per cent antiserum + streptodornase (1,000 U./ml.)	100

* Average of the counts of 3 experiments after 60 min. incubation at 37° C.

All the cells that still possessed a cell membrane, no matter how damaged they appeared, were counted.

Usually 500 cells were counted in each experiment.

[a] Ehrlich tumour cells 10^7/ml. washed with phosphate-buffer in saline, *p*H 7·4.

[b] Rabbit anti-Ehrlich serum.

[c] Anti-Ehrlich serum heated to 56° C. for 30 min.

[d] Tumour cells were incubated with antiserum for 15 min. at 37° C. before addition of human plasmin. Similar results were obtained with plasmin prepared from rabbit's serum.

The action of plasmin was found to be unspecific as both trypsin and papain were active in this respect. These *in vitro* findings raise the possibility that plasmin or other tissue proteolytic enzymes may be directly involved in cell destruction induced by the antigen-antibody reaction.

Work is in progress to determine what part, if any, these factors play in cellular damage *in vivo*.

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[1] Dale, H. H., *Acta Allerg.*, **5**, 191 (1952).
[2] Rocha, E., and Silva, M., *J. Immunol.*, **40**, 399 (1941).
[3] Ungar, G., Damgaard, E., and Hummel, F. P., *J. Exp. Med.*, **98**, 291 (1953).
[4] Hayashi, H., *Mie Med. J.*, **6**, 195 (1956).
[5] Ginsburg, I., *Brit. J. Exp. Path.*, **40**, 417, (1959).
[6] Salk, J. E., and Laurent, A. M., *J. Exp. Med.*, **95**, 429 (1952).
[7] Flax, M. H., *Cancer Res.*, **16**, 744 (1956).
[8] Goldberg, B., and Green, H., *J. Exp. Med.*, **109**, 505 (1959).
[9] Ratnoff, O. D., *J. Exp. Med.*, **87**, 199 (1948).
[10] Remmert, L. F., and Cohen, P. P., *J. Biol. Chem.*, **181**, 431 (1949).
[11] Pillemer, L., Ratnoff, O. D., Blum, I., and Lepow, I. H., *J. Exp. Med.*, **97**, 573 (1953).

Dietary Fats and Thrombosis

In order to study the influence of the fat content of food on the occurrence of thrombosis in the arterial and venous system, a group of 133 hospital patients aged 65–90 years were treated with a diet in which butterfat, margarine and lard were replaced by vegetable oils (unhydrogenated corn oil and soybean oil). The daily intake of vegetable oils was about 40 gm., or half the total fats. The time of treatment of the 133 patients ranged from 1 to 17 months (average 4·7 months) representing a total of 52½ years. For comparison, a control group of the same size and composition who received an ordinary hospital diet containing about 80 gm. animal fats (including about 40 gm. butter-fat, margarine and lard) was used. The occurrence of thrombosis and embolism in both groups was diagnosed clinically and post-mortem.

In the treated group only 4 cases of thrombo-embolism were observed: 1 case of myocardial infarction after 6 days of treatment, 1 case of myocardial infarction after 26 days of treatment, both verified by autopsy; 1 case of little stroke with transient aphasia of a few days' duration occurring after 30 days of treatment, and 1 case of pulmonary embolism after 4 months of treatment in a patient with bronchial cancer (verified by autopsy).

In the control group 15 cases of thrombosis were observed: 2 cases of thrombophlebitis (1 case complicated with pulmonary embolism and 1 case with thrombosis in the carotid artery), 2 cases of pulmonary embolism (both verified by autopsy), 5 cases of myocardial infarction (all typical, 2 verified by autopsy), 6 cases of cerebral thrombosis (3 verified by autopsy).

The difference in the occurrence of thrombosis in the two patient groups was statistically significant ($t = 5{\cdot}1$) and cannot be interpreted in terms of variations in age, weight and blood pressure in the groups.

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Interaction between the Plasma of the Loggerhead Turtle and Toxin of the Portuguese Man-of-War

THE loggerhead turtle, *Caretta caretta*, is reported to eat the Portuguese man-of-war, *Physalia physalis*[1-3]. Where *Physalia* are assembled in windrows, loggerheads are often seen at the surface with eyes closed, eating this siphonophore. *Velella*, an allied species, has also been identified from stomach contents of a turtle which was identified as a loggerhead[4]. *Physalia* is equipped with a singularly potent toxin[5]. This is contained in nematocysts which can penetrate a rubber glove and presumably even the chitinous exoskeleton of a crab's leg. As Barbour pointed out, the dense impenetrable skin of the face and jaws might protect a turtle from some of the obvious hazards of such a feast. However, since tentacles of active colonies have been observed to cling tightly to the skin, gloves, glass containers and even to the polished steel surface of a pair of scissors, it is difficult to explain the ability of the turtle to swallow, much less to digest and survive this meal. We believe that the acid of the stomach contents would inactivate the nematocyst and its contained venom virtually upon contact. The unconcern of the adult turtle with the toxic effects of the nematocysts might be due to the presence of protective antibodies in the blood acquired by contact with *Physalia* early in life. It is the object of this communication to report the results of experiments designed to test this hypothesis.

A 15-kgm. loggerhead turtle which had been in captivity less than one week was obtained from the Miami Seaquarium. The jugular vein was cannulated and the animal was bled. Sufficient potassium oxalate was added to the blood to bring the concentration to 0·2 per cent and the blood was cooled. Later the blood was centrifuged and the separated plasma was frozen at –20° C. without preservative. Toxin of *Physalia* was prepared according to procedures already described[5], with the following modifications: (1) Washed quartz sand and glass-distilled water were used for homogenization of the washed purified nematocysts. (2) After centrifugation, the material was mixed with 4/3 volumes of calcium phosphate gel prepared by the method of Kunitz[6] and was recentrifuged. The water-clear supernatant solution apparently contains all the toxic material. (3) This supernatant solution was lyophilized in tared 5-ml. capsules. These were sealed without admitting air and stored at –20° C. until used.

Fractions prepared in this way have regularly assayed at *LD*90 of 2·5–3·0 mgm./kgm. body-weight of pure strain, 18–25 gm. *C*57 black male mice when the mice are injected intravenously. The lyophilized material dissolves easily and completely. We assume that the entire weight of lyophilized material is toxic for purposes of estimating concentrations. It should be noted that if any significant fraction of the lyophilized 'toxin' were inactive then the active moiety would considerably exceed 2·5 mgm./kgm. in toxicity. The toxin is quite thermolabile and its activity decreases during manipulation.

In one experiment lyophilized toxin was dissolved in glass-distilled water to produce a concentration of 20 mgm./ml. One aliquot was diluted with 19 vol. of 0·9 per cent saline to a final concentration of 1 mgm./ml. (solution *A*, Table 1). The second aliquot was diluted with one volume of turtle plasma

Table 1

Solution	Toxin mgm. (dry weight)	Total diluent added Plasma (ml.)	Water (ml.)	0·9% Saline (ml.)	Final toxin concentration (mgm./ml.)
A	20	0	1	19	1
B	20	1	1	18	1
C	20*	1	1	18	1*
D	20*	1	1	0	10*

* Weight of toxin expected if it were all present in the supernatant solution (*C*) or in the precipitate (*D*).

and then further with 18 vol. of saline to a final toxin concentration of 1 mgm./ml. (solution *B*, Table 1). A precipitate formed when the plasma was combined with the toxin which did not dissolve when more saline was added. This precipitate was uniformly dispersed through the injection fluid and was included in the assay. The third aliquot was combined with an equal volume of turtle plasma and centrifuged to remove the precipitate. The supernatant liquid was assumed to contain all the toxin and was so diluted with saline as to bring the final toxin concentration to 1 mgm./ml. (solution *C*, Table 1). The precipitate was resuspended in a solution consisting of 1 ml. of turtle plasma and 1 ml. of glass-distilled water (solution *D*, Table 1). If all the toxic material were contained in the precipitate the concentration would have been 10 mgm./ml. Table 2 presents the results of assays of these solutions.

Table 2

Solution	No. mice injected	Dose-range (mgm./kgm.)	Surviving after 2 hr.
A	10	2·55–2·61	0
B	10	2·50–2·59	0
C	11	2·57–2·61*	0
D	10	47·0–56·2*	10

* Weight of toxin expected if it were all present in the supernatant solution (*C*) or in the precipitate (*D*)

Apparently all the toxicity originally present in solution *A* is still present in the toxin-plasma combination *B*. The precipitate which was separated and resuspended in saline *D* contained none of the activity. The supernatant solution *C* held all the toxicity of the original combination.

This experiment was repeated twice, with fewer mice, with entirely similar results. These results suggest that the loggerhead turtle lacks blood immune bodies which might be invoked to explain its apparent insensitivity to the toxin of *Physalia*.

Our interpretation does not exclude the possibility that the loggerhead might possess localized tissue antibodies. Phisalix[7] has shown that reptiles, and especially turtles, are less sensitive to the sting of bees than other vertebrates. It is possible that turtles are not susceptible to venoms of the type injected by *Physalia*.

We are grateful to Dr. Wilhelmina Dunning of the University of Miami for providing mice and to the Miami Seaquarium for the loggerhead turtle. These studies were supported by Grant *RG*-5272 from the U.S. Public Health Service, National Institutes of Health.

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[1] Babcock, H. L., *Proc. Zool Soc. Lond.*, A, 107, 595 (1937).

[2] Barbour, T., "Naturalist at Large" (Little, Brown and Co., Boston, 1943).

[3] Carr, A. F., "Handbook of Turtles" (Cornell University Press, Ithaca, New York, 1952).

[4] Murphy, R. C., *Copeia*, No. 2 (1914).

[5] Lane, C. E., and Eleanor Dodge, *Biol. Bull.*, 115, 219 (1958).

[6] Kunitz, M. J. *J. Gen. Physiol.*, 35, 423 (1952).

[7] Phisalix, Mme. M., *Bull. Mus. Nat. Hist. Paris*, 6, 166 (1934).

Elucidation of the Life-cycle of *Fasciola hepatica*

DR. B. DAWES, in his recent communication[1], has made an important contribution to our knowledge of the trematodes. In the interests of historical accuracy and in justice to a great nineteenth-century zoologist I would, however, like to point out that the life-histories of several trematodes, including that of *Fasciola hepatica*, were beautifully and accurately described by J. J. S. Steenstrup in his work "On the Alternation of Generations", translated into English and published as a Ray Society monograph in 1845, nearly forty years before the work of R. Leuckart and A. P. Thomas. Steenstrup solved one of the most difficult of all zoological problems, and his ideas on the alternation of generations were considered so revolutionary that G. Busk, his translator and editor, thought fit to write an apologetic introduction to assuage the anger of his readers.

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[1] *Nature*, 184, 1334 (1959).

I AGREE with H. S. Jefferies that historical accuracy is desirable and that the credit for scientific discovery should go where it is deserved. Parasitology owes much to the work of Steenstrup, who demonstrated the 'alternation of generations' which occurs in the lives of some invertebrate animals[1]. This expression was first used in 1819, however, by the Franco-German poet and naturalist, Louis Adelaide de Chamisso, in regard to the life-cycle of the pelagic tunicates known as salps, but the young Danish zoologists showed that it applies to the life-cycles of some cœlenterates, trematodes and other invertebrate animals as well. Valuable discoveries were made also by other zoologists, and some of these were mentioned in Chapter 16 and elsewhere in my book[2], and others have since been indicated by E. G. Reinhard, with special reference to the life-cycle of *Fasciola hepatica*[3].

In using the term 'cercaria', Steenstrup recognized both a discovery and an invention made in 1773 by O. F. Muller. The forms which he called 'nurses' were later named 'rediae' by F. de Filippi in honour of Francesco Redi who, in 1668, made all discoveries in this field possible by destroying the false doctrine of spontaneous generation. Such 'nurses' were first seen in 1818 by Ludwig Bojanus, who called them 'royal-yellow worms' and emergent cercariæ were first seen to encyst in 1807 by C. L. Nitzsch of Halle, who thus witnessed the formation of what Steenstrup later called 'pupae'. Some earlier discoveries made by two of C. A. Rudolphi's pupils did not enter into Steenstrup's considerations, though they concern the trematode life-cycle. In 1831, K. E. Mehlis described the hatching of 'ciliated embryos' from the eggs of trematodes, and in 1837, F. C. H. Creplin observed that such forms (miracidia) emerge from the eggs of *Fasciola hepatica*. Unluckily, Bojanus, Steenstrup and other persons mentioned here by name seem not to have made a study of the parasites of *Limnaea truncatula*, and for this reason did not observe the larval stages of *F. hepatica*. Steenstrup made a brief reference (5 lines) to the adult of this species and then turned to descriptions of echinostome and stylet cercariæ. He did not even mention the larval stages of the liver-fluke, and in fact he did not describe any trematode life-cycle in its entirety. According to Reinhard, the German parasitologist, Davis F. Weinland was the first person to suspect that the larval stages of the liver-fluke occur in *L. truncatula*. In 1875, he "told of finding cercaria-sacs" in the digestive gland of this snail, and he also noted that the cercariæ tended to leave water, which prompted him to suggest that they encysted on herbage and were the young of *Fasciola hepatica*. Thomas was apparently unaware of Weinland's discovery and conjectures, and his well-deserved success was the result of a methodical search for possible snail hosts in localities where sheep were liable to contract liver-rot. The quest was probably simplified by the knowledge that only fishes and molluscs were to be regarded with suspicion because the only animals known to harbour larval trematodes, but success called for all the resources of a most resourceful zoologist.

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[1] Steenstrup, J. J. M., "On the Alternation of Generations; . ." (The Ray Society, London, 1845) (first published 1842 in Copenhagen).

[2] Dawes, Ben, "The Trematoda" (Camb. Univ. Press, 1946, 1956).

[3] Reinhard, E. G., *Exp. Parasitol.*, 6, 208 (1957).

Differentiation induced as Season Advances in the Embryo-Gametophyte Complex of *Pinus nigra* var. *austriaca*, using Indole Acetic Acid

MAINTENANCE of growth in very young excised gymnosperm embryos in culture medium has been attempted by one of us with limited success. Indefinite growth was never achieved, and at death, differentiation was lacking other than that suggesting initiation of symmetry or polarity.

The present work on *Pinus austriaca* arises out of attempts to find qualitative evidence of physiological relationship in the region of the parental-embryonic threshold. Indole acetic acid, because of its acceptance as a growth-controlling agent, was added to the culture medium in the hope that it would stimulate differentiation during post-zygotic *in vitro* life.

Seeds were gathered at intervals during the year. Their testas were removed under aseptic conditions, and the naked gametophytes were placed on a sterile culture medium buffered to pH 6·0, and containing a known concentration of indole acetic acid. Various concentrations of indole acetic acid were sampled in the course of the experiments.

The gametophytes were harvested, some after two, others after four weeks *in vitro*.

Examination followed sectioning and staining in borax carmine and fast green.

Comparison of several gametophytes grown on an indole acetic acid medium in July immediately following fertilization showed in each case the presence of a meristematic layer crossing the gametophytic tissue. Position of the meristem varied from one example to another. It consisted of contiguous thin-walled cells resembling the fusiform initials of a vascular cambium.

Cells between the periphery of the gametophyte and the meristematic layer had undergone enlargement which produced local swelling. These cells contained little cytoplasm and only a few granules. Cells on the other side of the layer were much smaller, irregularly shaped, and contained a strikingly large number of granules which differed markedly in form from those normally present at corresponding positions in controls.

In many of these gametophytes, the parenchyma cells surrounding the archegonial jackets developed secondary thickenings on all their walls. Thus in these cells pits appeared on all the walls; they were large and circular.

Experiments performed in August, long after fertilization, showed neither the development of meristematic layers in gametophytic tissue nor the development of thickenings and pitting outside the archegonial jackets. Therefore, none of the phenomena arising in July examples occurred in the August ones.

In the older examples a different feature arose not seen in the July ones. At several scattered locations, cells one to three times the volume of neighbouring ones arose. These were thick-walled, lignified, pitted and often anisotropic in polarized light. Cytoplasm and nuclei were not visible in them.

Proembryo ontogeny for indole acetic acid *in vitro* cases differed from that for the controls in both the July and August examples. One cultured proembryo developed reticulate pitting in the walls of the suspensor cells. Others showed inhibited growth. Generally, on the basis that incidence of anomaly and differentiation was less frequent for embryo than for gametophytic tissue, the effect of the indole acetic acid *in vitro* conditions was judged to be less marked for the embryo system.

Experiments performed at the end of March with material collected at that time (nine months after fertilization) showed none of the previously described phenomena.

It may be concluded that: (1) In *Pinus austriaca*, as one example of an embryophyte, parental-embryonic environment shows a differential response to a factor stimulating differentiation. This differential suggests presence of a pattern of organization in which time and morphological events are significant and related. (2) In parental tissue, visual differentiation may be extensive; for example, meristem and massive activity near archegonia, or confined to single isolated cells, for example, lignified structures. In embryonic tissue the effect is always confined to single cells. (3) Indole acetic acid appears to be influential in initiating the differentiation and in effecting its course.

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Collection of Urine and Fæces from the Chicken

IN order to examine some factors which influence the intestinal absorption and urinary excretion of calcium and phosphorus in the chicken, a surgical method for the separation of urine and fæces has been developed which depends upon the successful 'exteriorization' of the distal end of the large intestine. This method, which has been employed principally in adult Rhode Island Red females, is equally suitable for adult capons and cockerels. Females operated on one year ago and fed practical rations have maintained normal behaviour, appetite, laying performance and live weight compared with non-operated controls. They have been employed for the daily collection of urine and fæces in various experiments, to be reported elsewhere, ranging from three to ten weeks duration.

Exteriorization of the distal end of the large intestine at a point on the abdominal wall has been previously reported by others. Rothschild[1] reviewed previous reports of techniques while contributing his own method, but mentioned no criteria of health and production in operated birds. Ariyoshi and Morimoto[2,3] described a technique that was afterwards used for nitrogen balance studies in cockerels over a period of six months and which necessitated the use of a special diet. Theirs is the only report of a long-term metabolic study on separated urine and fæces in the chicken.

The principal criticisms, therefore, of the previously described methods are: (1) they fail to provide such criteria of normality as behaviour, appetite and production; (2) there has been insufficient emphasis on, or attempt to explain, the post-operative complications and how to avoid them; (3) none has mentioned the quality of the fæces, which are invariably scant and fluid if there is any degree of stenosis or intestinal atony; (4) no long-term quantitative observations with laying females have been reported.

In carrying out the operation, general anæsthesia and full aseptic methods are essential. The terminal end of the large intestine as it joins the cloaca is ligated, incised, and exteriorized at a point half-way between the ischium and the abdominal mid-line approximately one and a half inches anterior to the ventral lip of the cloaca on the right side. The cloaca now forms a blind sac (except for the oviducal orifice in the female), and urine, which enters it from the ureters, is periodically expelled since the normal defæcation reflexes are retained. To collect the urine, a wooden frame covered with polythene sheet is placed below the wire floor of the cage. The polythene sheet slopes towards a central hole through which the urine flows into a vessel. The fæces, which are firm in consistency and in short lengths, pass out of the exteriorized bowel continually. To collect the fæces, a 100-ml. polythene beaker is inserted into a rubber cuff with a roll end sutured to the skin around the exteriorized bowel.

The production of a successful preparation depends upon the elimination of several post-operative complications. Any degree of stenosis due to inflammation, fibrosis, contraction of abdominal muscle or fascia and skin overgrowth around the exteriorized bowel must be avoided. Stenosis due to any of these factors always induces fæcal obstruction, constipation and intestinal atony. In order to overcome these complications, a thin-walled glass cannula, with approximately the same diameter, is inserted into the exteriorized bowel immediately after the operation. This cannula, through which fæces pass freely, is held in position by a polythene collar sutured to the skin immediately surrounding the exteriorized bowel. After two weeks, by which time contraction around the exteriorized bowl has terminated, the cannula is removed and fæces continue to pass quite freely. In some birds the skin immediately surrounding the bowel tends to overgrow the bowel orifice, but this difficulty is overcome by judiciously cutting it away. The birds remain clean and require little attention except in the course of experiments, when it is necessary to ensure that the fæces collector is securely attached.

I wish to express my thanks to members of the surgical unit at the Cambridge University School of Veterinary Medicine for their suggestions and criticisms, and I am indebted to the British Oil and Cake Mills, Ltd., for the provision of a research studentship which enabled this work to be carried out.

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[1] Rothschild, L., *Poultry Sci.*, **26**, 157 (1947).

[2] Ariyoshi, S., and Morimoto, H., Bull. Nat. Inst. Agric. Sci., No. 12, 87 (1956).

[3] Ariyoshi, S., and Morimoto, H., Bull. Nat. Inst. Agric. Sci., No. 13, 93 (1957).

Infection of Peas by Wilt Disease Fungi

A SERIOUS soil-borne wilt disease of peas has been shown to be caused by *Fusarium oxysporum* f. *pisi* (Linf.) Synder and Hansen, and *Fusarium oxysporum* Fr. emend. Synder and Hansen var. *redolens* (Wollenw.) Gordon[1,2]. Both these fungi attack the vascular system of the plant, and a typical internal symptom of the disease is an orange or brick-red discoloration of the vascular tissue.

During the examination of wilted pea plants which had been grown from seed in naturally infested soil it was noticed that, when the lowermost leaves began to wilt, which is the earliest recognizable stage, no discoloration could be found except in the cotyledonary vascular bundles. Furthermore, in plants which showed the curious 'half-leaf' wilt in which half of each leaf was wilted, only one of the two cotyledonary bundles was orange. Following these observations a series of isolations were made from the steles of plants showing various degrees of wilting. All plants with any sign of foot-rot (*Fusarium solani* f. *pisi*) were discarded, and as this fungus was not isolated from any of the experimental plants it is considered that the results in Table 1 refer to the wilt disease alone.

Table 1. DISTRIBUTION OF THE FUNGUS IN THE PLANTS AT VARIOUS STAGES OF WILTING

Leaves		Stem	Cotyledonary bundles		Hypocotyl	Root
Total	Wilted					
8	0	–	–	–	–	–
8	1	–	+	+	–	–
8	2	–	+	+	+	–
8	4	–	+	+	+	+
8	5	+	+	+	+	+
8	6	+	+	+	+	+
Completely wilted plant		+	+	+	+	+
'Half-leaf' wilt		+	+	–	+	+

An examination of Table 1 reveals that in the earliest recognizable stages of the wilt disease the fungus can be isolated from the cotyledonary bundles only; but as the wilt symptoms progressed so did the distribution of the fungus. Where the early stage of 'half-leaf' wilting was present the fungus was isolated from one cotyledonary bundle only. Consideration of the configuration of the vascular system of the pea plant[3] indicates that from a unilateral infection this 'half-leaf' wilting might be anticipated.

Microscopical examination of the distribution of the fungus within the plant confirms the foregoing observations. In 'half-leaf' wilt the fungus extends much farther up the vessels of the wilted side of the stem than of the non-wilted side, and in the root of an extensively wilted plant only two of the three points of the triarch stele, those corresponding to the cotyledonary bundles, contained the fungus.

It was concluded, therefore, that, in peas grown from seed in naturally infested soil, invasion of the host by the fungus is through the moribund cotyledons. Occasionally, where the root has had to curl around the seed before growing downwards, infection has been observed at the junction of an upper lateral root with the tap root. In plants in the first stages of wilting, infection at other points has been recorded in a very few cases, but it has always been associated with other damage, for example, by eelworms.

Field observations confirm these findings but, on land which has been too frequently cropped with peas, the root disease picture is often complicated owing to the multiplication of other soil-borne pests and diseases.

This investigation is continuing, and a more detailed report will be published later. I am indebted to the Agricultural Research Council for a grant enabling me to do this work, to Sir Joseph Hutchinson for laboratory and glasshouse facilities and to the National Agricultural Advisory Service for their co-operation.

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[1] Buxton, E. W., and Storey, I. F., *Plant Pathol.*, 3, 13 (1954).

[2] Buxton, E. W., *Trans. Brit. Mycol. Soc.*, 38, 309 (1955).

[3] Hayward, H. E., "The Structure of Economic Plants", 350 (The Macmillan Co., New York, 1938).

Arm Abnormality in *Octopus*

WHILE working through the cephalopod collections of the University of Miami Marine Laboratory I found a specimen which possessed an abnormal third left arm. The arm was divided into two branches. An unusual feature was the presence of a well-developed web between the two divisions as found uniting the bases of normal arms in this species.

Regeneration in cephalopods has been observed and reported by several workers but they are generally concerned with unipedal growth as a result of damage or autonomy. A. E. Verrill[1] reported on the regeneration of suckers in the giant squid, *Architeuthis harveyi*. Parona[2] described the occurrence of two, thin, short arms in the position of the first left arm of *Octopus vulgaris*; the presence of an extra arm between the second and third right arm in *Eledone aldrovandi*; and the vague mention of a divided arm in a specimen of *Eledone moschata*. Hanko[3] cited the occurrence of a split right fourth arm in a specimen of *Octopus vulgaris*. M. Lange[4] concerned herself with the histological processes and method of regeneration and only cited Hanko's 1913 work without further comment. Robson[5] mentioned the occurrence of regeneration, Lange's work, and autophagy. Sasaki[6] noted and figured a monstrosity of *Octopus vulgaris* in which the right fourth arm gave rise to 25 offshoots, all bearing suckers but with no indication of a web. He stated that he had observed such abnormality in two cases, both *Octopus vulgaris*. S. G. A. Jaeckel[7] reproduced Sasaki's figure.

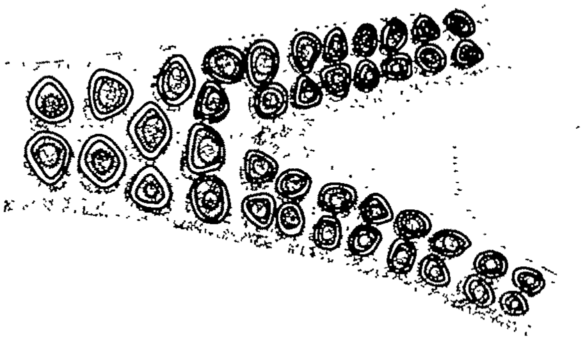

Fig. 1. Ventral view of the split-arm specimen of *Octopus briareus* Robson

The specimen (*Octopus briareus* Robson, 1929, U.M.M.L. 31 : 160) examined by me appears unique in the presence and extent of a web between the two extremities of the split third left arm. An existing web between extra arms has not been recorded previously or at least has escaped notice of the above-cited workers.

The specimen examined was a male, collected by Clench and Dickinson on June 7, 1956, at Big Pine Key, Florida. It was identified by means of the key and description given by Voss[8] for *Octopus briareus*. Basic measurements and indices are given in Table 1. Indices used are as defined by Pickford[9] and Voss[8].

Table 1. MEASUREMENTS (MM.) AND INDICES OF THE SPLIT-ARM SPECIMEN OF *Octopus briareus* U.M.M.L. 31 : 160 (MALE)

Mantle length	51·2
Mantle width	39 5
Mantle width index	77·1
Hectocotylized arm length	173·3
Ligula length	5 6
Ligula index	3·2
Arm order	3.2.4.1

The length of the third left arm (left ventro-lateral arm) as measured from the first basal sucker to the junction of the split is 94·3 mm. When the arm is viewed ventrally (sucker side up) the split sections measure 101·5 mm. and 103·5 mm., left and right side respectively.

The suckers are biserial in the normal fashion from the mouth to immediately below the junction. The second row of suckers, distally from the centre of the junction, is composed of three suckers across. The last row of suckers on the main stem has four suckers across the arm. On each of the divided sections the paired sucker arrangement found on the main stem is present. The web depth is 8·2 mm. between the two branches. A ventral view of the split arm is shown in Fig. 1. Measurements of the split arm are summarized in Table 2.

Table 2. MEASUREMENTS (MM.) OF THE SPLIT THIRD LEFT ARM

Arm length (to base of split)	94·3
Arm length of left branch	101 5
Arm length of right branch	103 5
Depth of web	12 1
Diameter of largest sucker	7·3

This type of growth does not appear to be the result of an arm injury. The web is shallow in this species and the presence of a web (11 per cent of the branched arm length) between the split sections would seem to indicate a mutant or an abnormality caused by injury to the embryo. No adaptive use for this development can be seen as the ligula is well developed and all other arms are intact, nor did the modification appear to be detrimental to the animal as it was in good shape and apparently healthy at the time of capture.

I wish to acknowledge the constructive criticism offered by Dr. G. L. Voss.

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[1] Verrill, A. E., *Trans. Conn. Acad. Sci.*, 5, Pt. 1 (1882).

[2] Parona, C., *Boll. dei Mus. di Zool. della Univ. Genova*, No. 96 (1900).

[3] Hanko, B., *Archiv Ent. Mech.*, 37, Heft 3 (1913).

[4] Lange, M., *J. Exp. Zool.*, 31 (1) (1920).

[5] Robson, G. C., "A Monograph of the Recent Cephalopoda", Pt. 1 (Brit. Mus., London, 1929).

[6] Sasaki, Madoka. *J. Coll. Agric. Hokkaido Imp. Univ.*, 20, Supp. No. 1 (1929).

[7] Jaeckel, S. G. A., "Cephalopoden Die Tierwelt der Nord- und Ostsee", Lief., 37, Teil 9 *bz*, Akademische Verlag. Geest und Portig (1958).

[8] Voss, G. L., Bull. Mar. Sci. Gulf. and Carib., 6, 2 (1956).

[9] Pickford, G. E., *Trans. Conn. Acad. Sci.*, 36, 3 (1945).

Delayed Birth in Marsupial Macropods—the Euro, the Tammar and the Marloo

THE first anecdotal evidence for delayed birth in macropods was given by Carson[1] in 1912. However, the phenomenon of delayed birth in the quokka (*Setonix brachyurus*) and the tammar (*Protemnodon eugenii*) was first investigated in detail by Sharman[2–4].

During a study of the euro (*Macropus robustus*) undertaken by the University of Western Australia we have found many delayed blastocysts lying free in the uteri while the females had 'joeys' in the pouch. The blastocysts were located by flushing out the uteri with physiological saline as described by Sharman[3]. Following this discovery an attempt was made to initiate a delayed birth in an experimental manner. A female euro obtained from the local zoological gardens was found to have a 28-gm. 'joey' in the pouch. This 'joey' was removed and the female isolated from further contact with males. 36 days later examination of this animal revealed a pouch young attached to the teat. Examination on the thirty-fourth day had shown no pouch young.

Previous work by Sharman[4] showed that tammars contain delayed blastocysts, and he has given anecdotal evidence which supported a delayed birth mechanism in this species. We have also produced a delayed birth pouch young in the tammar by removing a 0·70-gm. 'joey' and segregating the female from all males. Unfortunately, initial examination of the pouch was made 62 days after the removal of the first 'joey'.

During the course of the present work on marsupial macropods we have found blastocysts lying free in the uteri of two marloos (*Macropus rufus*) which both had 150-gm. 'joeys'. Apart from Carson's[1] initial observation this appears to be the only reported evidence for delayed birth in this species.

We acknowledge financial assistance from Western Australian University Research Grants and a generous grant for marsupial research from the Commonwealth Scientific and Industrial Research Organization executive.

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[1] Carson, R. D., *Proc. Zool. Soc.*, 234 (1912).

[2] Sharman, G. B., *Nature*, 173, 302 (1954).

[3] Sharman, G. B., *Aust. J. Zool.*, 3 (1), 56 (1955).

[4] Sharman, G. B., *Aust. J. Zool.*, 3 (2), 156 (1955).

ENTOMOLOGY

Time of Feeding in Relation to the Toxicity of a Stomach Poison to the Desert Locust

PEARSON[1] first showed that the stomach toxicity of dieldrin to fifth-instar hoppers (nymphs) of the desert locust (*Schistocerca gregaria* Forsk.) was increased by a period of starvation after treatment and suggested that this was due to variation in the rate of passage of the poison through the gut. We have confirmed and extended his observations and are directing attention to them because there appears to be little published information on the effects of starvation on the toxicity of stomach poisons to insects[2] although attention has been given to the effects of the diet fed to insects before they are used for experimental purposes[3].

When insects are fed with poisons applied by methods (for example, the poisoned sandwich method) other than those utilizing a poisoned medium there are usually time intervals between removal of the insects from their normal food, consumption of the poison, and resumption of normal feeding. In practice the insects are usually starved before an experiment, in order that they will readily accept proffered food and/or poison[2]. When the time-intervals are variable and of the duration of several hours they can affect the rate of passage through the gut of small quantities of food such as those that comprise a 'sandwich' containing poison.

A batch of fifth-instar nymphs, 4–7 days after moulting, was randomly divided into three groups. Two of the groups were deprived of food for 24 hr. Each insect in all three groups was then fed a small quantity of fluorescent powder ('phosphorescent powder, green', supplied by Rex Campbell, Ltd.) applied in gum to a small piece of grass. Immediately after eating this material two of the groups, including one of those previously starved, were then supplied with abundant fresh grass. The three groups of insects therefore received different treatments: group I was starved before dosing and fed after, group II was fed before and after, while group III was starved before and after. The fæcal pellets produced by each group in successive hours after ingestion of the indicator substance were counted. Each pellet was then broken up in about 1 ml. of water and examined in ultra-violet light, when any granules of the indicator present were clearly visible. The results obtained are shown in Table 1.

Table 1. TOTAL NUMBER OF FÆCAL PELLETS AND NUMBER CONTAINING INDICATOR AT DIFFERENT TIMES AFTER DOSING (PER GROUP OF 10 HOPPERS)

Treatment group	Hours after dosing				
	0–1	1–2	2–3	3–4	5–6
I	7/19	23/31	14/30	18/32	5/36
II	*	2/22	8/18	15/16	16/30
III	0/3	0/3	0/4	3/3	7/7

* Owing to difficulty in making these insects feed, no estimate can be given for this period.

It will be seen from Table 1 that the proportion of pellets containing the indicator depends on the treatment given. However, the quantity of indicator substance in each pellet was very variable, and rough quantitative estimates were made visually of the amount lost per hour. It was estimated that 80 per cent was lost in less than 2 hr. in group I, in 4–5 hr. in group II, and in 7 hr. in group III.

In further experiments the same range of doses of the insecticide dieldrin was fed to each of two groups of hoppers which had been starved for a period of 24 hr. The insecticide was applied to a small portion of a blade of grass, and within 15 min. of eating this the hoppers of one group were supplied with abundant freshly cut grass. No deaths occurred within five days among the 56 insects in this group. The second group was kept without food for a further 24 hr. after dosing and a total of 48 deaths occurred during the next five days from the 55 insects that were dosed. These results corresponded to *LD*50's of $\gg$ 4·8 and 0·79 μgm./gm. respectively. In the second experiment similar results were obtained, that is, *LD*50's of $\gg$ 4·8 and 0·95 μgm./gm.

Pearson's toxicity results clearly fit in with these findings. His normal treatment most nearly followed group I above, since his insects "were not fed on the previous evening" but were fed immediately after dosing, while his starvation treatment was equivalent to group III. As a result of the different treatments the resistance of the insects fed immediately after dosing was increased 1·5–2·8 times.

It is interesting to note that what might appear to be a convenient laboratory procedure may yield very variable results. For example, locusts may be deprived of food for 24 hr. before a stomach toxicity experiment is due to be carried out and then given the poisoned food. Later, when convenient, they are supplied with abundant grass ; if this occurs immediately after the poison is consumed, the toxicity will be very much less than if there is a delay of a few hours, as may well occur when a large series of individuals is being treated.

In the field, Ellis and Ashall[4] found that in some cases hoppers exhibited a noticeable diurnal rhythm in feeding, there being least feeding during the 6 hr. after dawn, and most between 1600 and 2200 hr. It is thus probable that when dieldrin is sprayed on vegetation, a technique being used on an increasing scale[5] for locust control, its toxicity will vary with the time of day at which it is eaten.

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[1] Pearson, A. J. A., Proc. Fourth Internat. Congr. Crop Protection, Hamburg (1957).

[2] Busvine, J. R., "Techniques for Testing Insecticides" (London, 1957).

[3] Potter, C., and Way, M. J., in "Methods of Testing Chemicals on Insects", ed. Sheppard, H. H., 1 (Minneapolis, 1958).

[4] Ellis, P. E., and Ashall, C., *Anti-locust Bull.*, No. 25 (1957).

[5] Rainey, R. C., *J. Sci. Food and Agric.*, 9, 677 (1958).

Fertilization of Eggs of the Desert Locust by Spermatozoa from Successive Copulations

In the desert locust (*Schistocerca gregaria* Forsk.) copulation occurs frequently, although once fertilization has taken place further copulation appears to serve mainly as a stimulant to egg-laying and to be unnecessary for renewed fertilization[1]. An interesting problem arises as to whether, after a second copulation, eggs are fertilized by sperm from the first or the second copulation. The following experiment was devised, using an albino stock of *Schistocerca*. It has been shown previously that crosses between albino and normal adults show a Mendelian pattern of inheritance, in which albinism is recessive[2].

Four pure albino females were kept in isolation in cages, and when they were sexually mature an albino male was introduced into each cage for 24 hr. and then removed. Subsequent oviposition produced averages of 60–80 young locusts per egg pod, and these included both males and females, showing that copulation had taken place. After each albino female had laid two egg pods, from which only albino hatchlings resulted, a normal-strain male was placed with each of the females and then removed after 24 hr. All subsequent egg pods produced young locusts that were normal in appearance, to the almost complete exclusion of albinos. In fact, out of 1,033 eggs that hatched, 1,028 were normal and five were albino. In a further experiment, the same procedure was adopted except that the albino female was fertilized first by a normal male and then by an albino ; the progeny of the first two pods were all normal in appearance, but after the second copulation all, without exception, were albino.

It is known that spermatozoa can fertilize eggs at least ten weeks after the last copulation[1], but from the present experiments it appears that a second injection in some way prevents further use of the previous spermatozoa. This is evidently not due to competition between a more and a less virile strain, as it occurred whether the albino or the normal male was used second. Whether the first spermatozoa were inactivated in some way, or even killed, by the second, or the second spermatophore mechanically prevented the first spermatozoa from reaching the eggs, cannot be deduced from these observations.

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[1] Norris, M. J., *Anti-Locust Bull.*, No. 18 (1954).

[2] Hunter-Jones, P., *Nature*, 180, 236 (1957).

OCEANOGRAPHY

Manganese Deposition on the Shells of Living Molluscs

Recently, there has been a renewal of interest in manganese deposition on the sea bed, particularly that brought about by biological rather than inorganic processes[1–6]. During the past fifty years[1–14] various theories have been put forward concerning the method of manganese deposition, yet the only measurements on the rate of deposition are those of Petterssen[11,12], and Buttler and Houtermans[13] on manganese nodules of deep-sea origin. These authors give a rate of the order of 1 mm. in 1,000 years.

During a survey of the sublittoral fauna of the Clyde sea area from 1949 onwards, it has been observed that the shells of many living molluscs are encrusted with a black deposit. In particular, this was the case of molluscs taken from muds and sandy-muds at depths of 15–105 fathoms. Although this has been recorded before[7–9,15], past analyses were limited to nodules, and deposits on stones and empty shells. While many species of living molluscs in the Clyde have manganese deposited on their shells, it is significant that deposits are more common and much heavier on shells with a dull, matt periostracum than on those with a glossy periostracum.

Yellow, and olive-coloured shells seem to be most affected, although deposits occur on shells of other colour. Thus species of *Nucula* clearly show the difference between glossy and matt surfaces, for while matt *N. sulcata* and *N. nucleus* may be completely covered with the black deposit (none of the older shells of many hundreds examined is free of deposit), glossy *N. turgida* and *N. tenuis* from the same substratum and of the same age rarely show any deposit. When present in the two latter species the deposit is confined to a small, thin film at the umbo. Although the list is not exhaustive, other species noted for their black deposit are *Corbula gibba*, *Venus casina*, *Nuculana minuta*, *Cyprina islandica*, *Mysella bidentata*, *Glycymeris glycymeris*, the underside of *Pecten maximus* and *Chlamys septemradiata*, *Natica alderi*, *Neptunea antiqua* and the tips of *Dentalium entalis*. All these species live at or close to the surface of the bottom sediment.

Analysis confirms that a high percentage of manganese is present in the deposits on these shells, far more than in the bottom sediment in which they live (Table 1). The percentage of manganese is close to that estimated for nodules from the same area. As figures for the rate of growth of *Nucula* species are available[16] micrometer measurements of the thickness of the deposit at the umbo give some idea of the rate of deposition. Shells of *N. sulcata* and *N. nucleus* less than 5 years old have a thin coating that is not easily measured; but the deposit on the dorsal side of shells more than 8 years old can be easily flaked off and its thickness determined. The results are shown in Fig. 1. It will be seen that, exceptionally, a deposit 1·27 mm. thick can be laid down in 12 years, but most fall in the range 0·30–0·70 mm. in 17 years (1 mm. in 25–60 years). It is possible that some of the deposit may be rubbed off by the movements of the *Nucula*, although the umbo, which is always affected, probably projects above the surface of the mud and so avoids friction with the substratum. Maximum deposition of manganese occurs on shells and non-living loci at the surface of the bottom sediment[6]. The deposit on living molluscs is remarkably compact and hard, perhaps suggesting origin from biological activity rather than the adsorption of precipitated particles. The possible origin of the Clyde nodules and deposits from streams flowing off manganese and iron-rich bogs cannot be neglected, although a higher percentage of manganese in the bottom sediments might be expected. Such an origin was discussed by Murray and Irvine[9]. The figures given above suggest that the largest nodules (approx. 6 mm. diam.) in a sample taken from the Minard Narrows (L. Fyne) are 75–190 years old. This is in contrast with 3,000 years for deep-sea nodules of the same size.

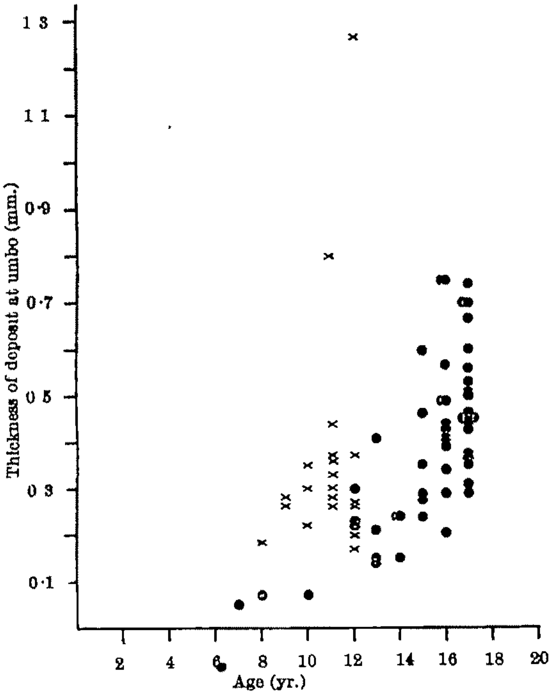

Fig. 1. Rate of manganese deposition on the shells of living *Nucula nucleus* (×) and *N. sulcata* (●)

Table 1

Locality	Manganese (per cent)
Nodules from Minard Narrows (Clyde)	18·7
Nodules from other sources (Goldberg) (ref. 3)	10·8–24·15
Substratum Arran Deep (Clyde) 60 fm.	0·1
Substratum North Sea 30 fm.	0·1
N. sulcata, Cumbrae Deep (Clyde), 60 fm.	8·5
N. sulcata, Arran Deep (Clyde), 60 fm.	17·9
Astarte montagui, Inverkip Bay (Clyde), 25 fm	11·5

I am indebted to Dr. A. Gilchrist of the Metallurgy Department, King's College, Newcastle upon Tyne, who kindly carried out the manganese estimations.

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[1] Graham, J. W., and Cooper, S. C., *Nature*, **183**, 1050 (1959).

[2] Graham, J. W., *Science*, **129**, 1428 (1959).

[3] Goldberg, E. D., *J. Geol.*, **62**, 249 (1954).

[4] Goldberg, E. D., and Arrhenius, G. O. S., *Geochim. Cosmochim. Acta*, **13**, 153 (1958).

[5] Dorff, P., "Die Eisenorganismen" (2) "Biologie des Eisen-und Mangankreislaufs" (1935).

[6] Murray, J., and Renard, A. F., "Deep Sea Deposits", Challenger Rept (1891).

[7] Buchanan, J. Y., *Trans. Roy. Soc. Edin.*, **36**, 459 (1891).

[8] Irvine, R., and Gibson, J., *Proc. Roy. Soc. Edin.*, **18**, 54 (1891).

[9] Murray, J., and Irvine, R., *Trans. Roy. Soc. Edin.*, **37**, 721 (1894).

[10] Correns, C. W., Deutsche Atlantische Exped. *Meteor* 1925–1927, Wiss. Erg., **3**, 298 (1939).

[11] Pettersson, H., *Göteborgs VetenskSamh. Handl.*, Ser. B, **3** (1945).

[12] Pettersson, H., *Göteborgs VetenskSamh. Handl.*, Ser. B, **2** (1943)

[13] Buttler, H. V., and Houtermans, F. G., *Naturwiss.*, **37**, 400 (1950).

[14] Buser, W., Int. Oceanogr. Congr. Preprints, **962** (1959).

[15] Chumley, J., "The Fauna of the Clyde Sea Area" (1918).

[16] Allen, J. A., *J. Mar. Biol. Assoc. U.K.*, **33**, 457 (1954).

PSYCHOLOGY

Scoring of a 'Steadiness' Test

A TEST of 'steadiness' has been used in this Department as a class experiment, in order to illustrate the importance of the 'double blind' principle in the design of controlled clinical trials. The apparatus of Seashore and Adams[1] was chosen. Steadiness is measured as the ability to hold a metal stylus in position for a fixed period within holes of graduated diameters, drilled in a metal plate, without allowing the stylus to come into contact with the sides of the hole. An index of steadiness is obtained by multiplying the contact times for each hole by arbitrary factors related to the hole diameters, and summing the result.

The timer used in this experiment was constructed from 'Add-on' dekatron units (Ericsson Telephones, Ltd., Beeston, Nottingham). A time signal (100 p.p.s.) is fed into the counting device through the input selector. With the 'count' key fixed in the reset position, the counter is set up for automatic starting and resetting through the socket of the terminal dekatron unit, the connexions being made by means of a 12-pin miniature Jones plug. The counter starts operating when the terminal *PLA*9 is earthed and is reset by removing the earthing temporarily from *PLA*10. In the 'start' circuit there is a Post Office 'Uniselector' driven by 40 V. d.c. pulses from a time clock. The contacts in the 'Uniselector' are connected in such a way that, after a 2·5-sec. interval, the 'start' circuit is completed for a period of 10 sec. The 'start' circuit also contains a high-speed relay, which is actuated by a 40-V. d.c current, 'made' when the stylus touches the sides of the hole. The relay closes the 'start' circuit and the counter operates. When the stylus is not touching the edge of the hole, the relay contacts are open, and the counter stops. Readings are thus obtained directly (in units of 0·01 sec.) of the time in a 10-sec test period during which the stylus is in contact with the hole.

Experiments were carried out on 58 students aged 19–32, 54 of whom were aged 23 or less. The test procedure was as follows. Students were divided into pairs, one acting as subject and the other working the apparatus. The controlled testing situation for the stationary position recommended by Seashore and Adams[1] was adopted, with the exception that the apparatus allowed each subject a 2·5-sec. interval to become accustomed to the operation of the timer. A test run consisted of four replicate trials, each comprising one reading apiece in the five holes the dimensions of which are given in Table 1,*a*. After a control run, the subject swallowed a tablet, which might contain either a stimulant drug or a placebo. An hour later a further test run was carried out. In the conditions of the experiment, a 'placebo effect' could be demonstrated, contact times being significantly decreased ($P < 0{\cdot}001$) at the second test run. No learning or fatigue effects during either run could be demonstrated.

Table 1. CORRECTION FACTORS FOR A 'STYLUS AND HOLE' STEADINESS TEST

Results of 232 observations on 58 subjects. To obtain a standardized score the contact times for each hole are multiplied by the appropriate correction factor and the results are summed

(*a*) Hole diameter (1/64 in.)	12	11	10	9	8
(*b*) Mean contact time in a 10 sec. test period (0·01 sec)	224·9	97·7	46·0	29·2	20·8
(*c*) 1/(*b*) × 100	0·445	1·023	2·174	3·425	4·808
(*d*) Correction factors (Spaeth and Dunham, ref. 2)	0·5	1	5	10	20
(*e*) Correction factors based on (*c*)	1	2	4	7	10

Mean contact times for each of the five holes were calculated from the control runs, comprising 232 readings in each hole (see Table 1,*c*). Some arbitrary assumption is necessary to render comparable the five respective means. At first sight it would appear simplest to use multiplication factors proportional to the reciprocals of the observed mean contact times so that, after correction, the readings from each hole would give, on the average, the same numerical result. The correction factors recommended by Spaeth and Dunham[2] (Table 1,*d*) do not obey this relation. An alternative series of integer factors which possesses approximately the desired property is given in Table 1,*e*. It is possible, however, that investigation of the wave form of upper limb tremor in relation to the geometry of the apparatus might show that some other basis for correcting the reading was preferable.

I wish to thank the students in the Pharmacology and Statistics Departments of the University of Sheffield who took part in the experiments, Mrs. W. M. Wright for help and statistical analyses and Dr. D. R. Wood for advice in constructing the apparatus.

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[1] Seashore, R. H., and Adams, R. D., *Science*, 78, 285 (1933).

[2] Spaeth, R. A., and Dunham, G. C., *Amer. J. Physiol.*, 59, 467 (1922).

STATISTICS

Fitting ν Linear Relations to n Variables all liable to Error

IN a recent communication, B. E. Kelly[1] has directed attention to W. E. Deming's sadly neglected monograph "Statistical Adjustment of Data" (1943), and comments that Deming's procedure leads to the solution found by P. A. Wayman[2] for the best-fitting straight line to a series of observations of two variables both subject to variable error. This is not correct: Deming neglected the variation of his weighting factor W_i with the slope of the fitted line, and his result (p. 180) is a weighted regression. We have re-examined the problem of multivariate linear relations: when both variables are subject to independently variable errors we obtain a result the bivariate case of which agrees with Wayman's (which so far as we can find had not previously been described); when the ratio of the standard deviations of error in the several variables is constant for all sets of measurements, we find that for n variables connected by ν linear relations the best-fitting $(n - \nu)$-flat is defined by the ν latent vectors of the covariance matrix corresponding to its ν smallest latent roots; and we have been able to find a procedure suitable when not all the variables were measured at every point (a situation very common in studies of the correlation of chemical composition and physical properties of minerals). Certain non-linear relations of mineralogical importance are under investigation.

D. E. Barton and C. L. Mallows[3], discussing methods of curve-fitting other than least-squares, mention J. Berkson's method[4] of the 'controlled variable'. This method is based on the assumption that if in a series of experiments we adjust one variable as nearly as we can to a series of pre-selected 'control' values (one measurement at each) it will be proper to assume that the error of measurement is independent of the observed value, but that if no such 'control' is attempted the error of measurement is, necessarily, independent of the true value and not of the observed value; we are unable to find any justification for this postulate, and believe this procedure to be wholly invalid.

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[1] Kelly, B. E., *Nature*, 184, 1086 (1959).

[2] Wayman, P. A., *Nature*, 184, 77 (1959).

[3] Barton, D. E., and Mallows, C. L., *Nature*, 184, 1086 (1959).

[4] Berkson, J., *J. Amer. Statist. Soc.*, 45, 164 (1950). Cf. Lindley, D. V., *Biometrika*, 40, 47 (1953).

FORTHCOMING EVENTS

(*Meetings marked with an asterisk * are open to the public*)

Monday, February 1

Society of Engineers (at the Geological Society, Burlington House, Piccadilly, London, W.1), at 5.30 p.m.—Mr. E. G. Massey Collier: Presidential Address.

Society of Chemical Industry, London Section (at 14 Belgrave Square, London, S.W.1), at 6.30 p.m.—Mr. K. D. B. Johnson: "Industrial Separation of Isotopes".

Tuesday, February 2

Plastics Institute (at the Wellcome Building, 183–193 Euston Road, London, N.W.1), at 4 p.m.—Symposium on "Preplasticizing in Injection Moulding".

Institution of Electrical Engineers, Measurement and Control Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. P. Vigoureux: "Development of the Formulæ of Electromagnetism in the M.K.S. System".

University of London (at Imperial College of Science and Technology, South Kensington, London, S.W.7), at 5.30 p.m.—Prof. F. C. Tompkins: "Superficial Chemistry and Solid Imperfections".* (Inaugural Lecture.)

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Dr. Paul Fourman: "Magnesium Metabolism in Man".* (Seventh of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on February 4, 9, 11, 16, 18, 23, 25 and March 1).

University of London (at the Middlesex Hospital Medical School Mortimer Street, London, W.1), at 5.30 p.m.—Dr. Honor B. Fell "The Experimental Transformation of Cells"*.

British Interplanetary Society (at Caxton Hall, Westminster, London, S.W.1), at 7 p.m.—Mr. A. Wilson: "The Use of Reinforced Plastics in Rockets".

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Mr. H. H. Pearcey: "Suppression of Shock-induced Separations by Boundary Layer Control".

Wednesday, February 3

Royal Society of Arts (at John Adam Street, Adelphi, London, W.C.2), at 2.30 p.m.—Mr. Clifford Paine: "Modern Dyes" (Sir William Jackson Pope Memorial Lecture).

Royal Statistical Society (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5 p.m.—Mr. R. L. Plackett: "Models in the Analysis of Variance".

Society of Chemical Industry, Heavy Organic Chemicals Group (at 14 Belgrave Square, London, S.W.1), at 6 p.m.—Mr. D. J. Hadley: "The Preparation and Use of Nitriles".

Society for Analytical Chemistry (joint meeting with the Oils and Fats Group of the Society of Chemical Industry, at the Chemical Society, Burlington House, Piccadilly, London, W.1), at 7 p.m.—Meeting on "Spectroscopic Investigation of Fats".

Thursday, February 4

Royal Society (at Burlington House, Piccadilly, London, W.1), at 10.30 a.m.—Discussion on "The Results of the Royal Society I.G.Y. Expedition" opened by Sir Graham Sutton, F.R.S.

Physical Society, Optical Group (at Imperial College, London, S.W.7), at 2.30 p.m.—Dr. H. C. King: "The London Planetarium"; Mr. G. M. Sisson: "Present Plans for the Isaac Newton Telescope".

Institution of Naval Architects (at 10 Upper Belgrave Street, London, S.W.1), at 4.45 p.m.—Scientific Papers.

University College (in the Physiology Theatre, Gower Street, London, W.C.1), at 5 p.m.—Dr. Elizabeth Work: "The Mucopeptides of Bacterial Cell Walls".* (First of two lectures in Biochemistry. Further lecture on February 11.)

Institute of Refrigeration (at the Institute of Marine Engineers, The Memorial Building, 76 Mark Lane, London, E.C.3), at 5.30 p.m.—Mr. J. M. Buist and Mr. R. Hurd: "Polyurethane Foams—Their Contribution to Heat Insulation".

Institution of Electrical Engineers (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. J. H. Gridley: "The Shielding of Overhead Lines against Lightning".

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. E. B. Verney: "Some Aspects of the Renal Excretion of Water and Electrolytes".* (Eighth of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on February 9, 11, 16, 18, 23, 25 and March 1.)

Friday, February 5

Institution of Electrical Engineers, Medical Electronics Discussion Group (at Savoy Place, London, W.C.2), at 5.30 p.m.—Discussion on "Computers in Medical Use" opened by Dr. R. A. Buckingham and Dr. J. M. Tanner.

Society of Dyers and Colourists (at the Royal Society, Burlington House, Piccadilly, London, W.1), at 6 p.m.—Mr. L. Cotton and Mr. J. Wardle: "Dyeing and Finishing of 'Courtolon' Nylon in Relation to Its Yarn Properties".

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Prof. J. F. Danielli, F.R.S.: "Some New Approaches to Inheritance and Evolution" (The Woodhull Lecture).

Saturday, February 6

London County Council (at the Horniman Museum, London Road, Forest Hill, London, S.E.23), at 3.30 p.m.—Miss Maitland Howard: "Cats and Magic".*

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Assistant Lecturer in Pure Mathematics, and an Assistant Lecturer in Applied Mathematics in the Department of Mathematics; and an Assistant Lecturer in Applied Mathematics in the Department of Theoretical Mechanics—The Registrar, The University, Nottingham (February 6).

Chemist and Bacteriologist (with an honours degree in chemistry, member of the Royal Institute of Chemistry, and extensive experience of water treatment and examination)—Fylde Water Board, Sefton Street, Blackpool (February 8).

Librarian (with a degree or recognized qualification in librarianship, experience in a special library, preferably serving a research organization), to take charge of the Library at the Central Electricity Generating Board's Research Laboratories at Leatherhead, Surrey—The Personnel Officer, Central Electricity Generating Board, 24/30 Holborn, London, E.C.1, marking envelope "Confidential Ref. N/9" (February 8).

Assistant Lecturer in British Archæology—The Registrar, The University, Leicester (February 12).

Assistant Lecturers in Chemistry, Physics and Zoology—The Registrar, The University, Leicester (February 12).

Assistant Librarian (man or woman graduate)—The Registrar, The University, Leicester (February 12).

Lecturer or Assistant Lecturer (with a good honours degree and experience in teaching and research) in Mathematics at the University of Malaya, Kuala Lumpur—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (London and Malaya, February 12).

Principal of Cornwall Technical College, Camborne—The Secretary for Education, County Hall, Truro, Cornwall (February 12).

Lecturer in Education Psychology—The Registrar, The University, Liverpool, quoting Ref. CV/T (February 13).

Research Assistant in the Postgraduate Department of Applied Mechanics—The Registrar, The University, Sheffield (February 13).

Research Assistant (with a good degree, and preferably a higher degree in physics, chemistry, engineering or ceramics) in the Houldsworth School of Applied Science, to work on the high-temperature properties of certain ceramics of potential interest as engineering materials—The Registrar, The University, Leeds 2 (February 13).

Gulbenkian Research Fellow, to investigate certain aspects of the teaching of physics or chemistry in schools—The Registrar and Secretary, The University, Bristol (February 15).

Lecturer or Demonstrator in Chemical Pathology, for duties divided equally between original investigation and supervision of the hospital laboratory—The Registrar, The University, Leeds 2 (February 15).

Reader in Applied Mathematics—The Clerk to the Governors, Chelsea College of Science and Technology, Manresa Road, London, S.W.3 (February 15).

Editorial Assistant, Assistant Experimental or Experimental Officer, in the Publication Section, Road Research Laboratory, D.S.I.R., Harmondsworth, Middlesex, for work involving the editing and preparing for publication articles to technical journals, official booklets issued by the Laboratory, etc.—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting A.21/0A (February 19).

Junior Lecturer (preferably with the ability to share in the teaching of basic courses in psychology) in Education—The Registrar, Trinity College, Dublin (February 22).

Chair of Zoology at Westfield College—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 25).

Lecturer in Petrology in the Department of Geology—The Secretary, The University, Edinburgh (February 27).

Turner and Newall Research Fellows in Engineering, Inorganic Chemistry, Physics and allied subjects—The Registrar, The University, Manchester 13 (February 27).

Assistant Lecturer or Lecturer in Animal Husbandry in the Faculty of Veterinary Science—The Registrar, The University, Liverpool, quoting Ref. CV/T (February 29).

Leverhulme Postgraduate Fellows (3), 2 in the Faculty of Science and 1 in the Faculty of Engineering—The Registrar, The University, Liverpool, quoting Ref. CV/T (March 1).

Chair of Biochemistry at King's College, Strand, London, W.C.2—The Academic Registrar, University of London, Senate House, London, W.C.1 (March 2).

Lecturer or Assistant Lecturer in Botany at the University College of the West Indies—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 5).

Lecturer or Assistant Lecturer in Physics—The Registrar, The University, Southampton (March 5).

Imperial Chemical Industries Research Fellows in Chemistry or Physics—The Registrar, The University, Hull (March 7).

Associate Professor of Mathematical Statistics at the University of New England, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, March 12).

Imperial Chemical Industries Research Fellows in Biochemistry, Chemistry, Engineering, Pharmacology or Physics—The Secretary, The University, Aberdeen (March 12).

Reader or Senior Lecturer in Agricultural Chemistry, and a Lecturer in Animal Physiology in the Faculty of Agriculture, Makerere College, University College of East Africa—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 15).

Senior Lecturer (graduate of a British university or equivalent qualifications and experience appropriate to the post) in Physics; and an Assistant Lecturer (with a good honours degree in physics and preferably some experience of teaching and research) in Physics at the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, March 15).

Lecturer (preferably with a special interest in the field of transition elements or of complexes) in Inorganic Chemistry—The Secretary, The Queen's University, Belfast (March 18).

Lecturer and an Assistant Lecturer (preferably with a research interest in the application of mathematics to some engineering subject) in Engineering Mathematics—The Secretary, The Queen's University, Belfast (March 30).

Isaac Wolfson Research Fellow (suitably qualified graduate aged 24–35, either British subject (from any Commonwealth country) or Israeli citizen) in Metallurgy—The Principal, St. Edmund Hall, Oxford (April 23).

Associated Electrical Industries Fellow for Research in the Department of Physics—The Secretary-General of the Faculties, The Old Schools, The University, Cambridge (April 30).

Assistant Lecturer in the Department of Nutrition—The Secretary, Queen Elizabeth College (University of London), Campden Hill Road, London, W.8.

Associate Dean of the School of Art and Sciences; Professor of Electrical Engineering; Professor of Engineering Physics; and a Professor of Mechanical Engineering, at the Middle East Technical University, Ankara—Room 139, Foreign Office, London, S.W.1.

Chemist (with a good honours degree in agricultural chemistry or chemistry and a knowledge of field experimentation) in the Department of Agriculture, Uganda, for research on soil fertility problems connected with tropical crop rotations, manures and fertilizers—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.63/9/09/T.

Lecturer (well qualified physicist, with research, industrial or teaching experience) in the Science and Mathematics Department—The Principal, Rugby College of Engineering Technology, Eastlands, Rugby.

Lecturer (with a degree in chemical engineering or metallurgy) in the Department of Metallurgy, to teach the engineering principles of extraction metallurgy, particular attention being given to the effects of heat and mass transfer and fluid flow on the design and operation of metallurgical processes—Prof. F. D. Richardson, Department of Metallurgy, Imperial College of Science and Technology, Prince Consort Road, London, S.W.7.

Lecturer (with an honours degree in chemistry, with postgraduate experience of academic or industrial research) in Organic Chemistry—The Secretary, Sir John Cass College, Jewry Street, London, E.C.3.

Master to teach Physics in the Upper School, including Sixth Forms—The Headmaster, Ellesmere College, Shropshire.

Physicist, Senior grade (with at least five years experience in radiotherapy physics)—The Group Secretary, Mount Vernon Hospital, Northwood, Middlesex.

Research Assistant (preferably with an honours degree in zoology or medical sciences, and a knowledge of German) in the Department of Anatomy, for work mainly on the central nervous system—The Secretary, University College, Gower Street, London, W.C.1.

Research Entomologist (male, with a good honours degree in natural science, or a degree in agriculture with honours in entomology, and experienced in entomological research) in the Department of Agriculture, Nyasaland, to assist with investigation of cotton pest problems in the Federation of Rhodesia and Nyasaland—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD./63/4/016/T.

Research Fellow (with a good degree in electrical engineering, physics or chemistry and some research experience of vacuum systems) in the Department of Electrical Engineering—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1.

Scientific Assistant, to assist an entomologist developing a technique to control *Culicoides impunctatus* in Scotland—R. Parish, B.Sc., Department of Zoology, University of Edinburgh, West Mains Road, Edinburgh 9.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

Manchester Literary and Philosophical Society. Memoirs and Proceedings, Vol. 101, 1958–1959. Pp. 121+xii. (Manchester: Manchester Literary and Philosophical Society, 1959.) 31*s*. 6*d*. [312

De Havilland Propellers Limited. The De Havilland Transistor Curve Tracer. Pp. 4. Publication 1076: The De Havilland Portable Automatic Multi-core Cable Tester. Pp. 6. General Data. Pp. 7. Publication 5078: De Havilland Electromechanical Torque Motors—General Information. Pp. 8. (Hatfield: De Havilland Propellers Limited, 1959.) [312

Imperial College of Science and Technology. Inaugural Lectures, 1956–57 and 1957–58. Pp. ii+134+24 plates. (London: Imperial College of Science and Technology, 1959.) [312

Department of Scientific and Industrial Research and Fire Offices' Committee. Flame-Retardant Building Materials. Pp. iv+8. (London: H.M. Stationery Office, 1959.) 1*s*. net. [312

Privy Council. Medical Research Council Memorandum No. 38: Effect of Small Doses of Alcohol on a Skill Resembling Driving. By G. C. Drew, Dr. W. P. Colquhoun and Hazel A. Long. Pp. vi+108. (London: H.M. Stationery Office, 1959.) 7*s*. 6*d*. net. 312

British Museum (Natural History). Instructions for Collectors No. 11: Fossils, Minerals, and Rocks. Seventh edition. Pp. 16. 1*s*. Bulletin of the British Museum (Natural History). Historical Series. Vol. 2, No. 1: Darwin's Journal. Edited by Sir Gavin de Beer. Pp. 1–21. 8*s*. Entomology. Vol. 8, No. 5 The Walker Types of Fruit Flies (Tephritidae-Diptera) in the British Museum Collection. By D. Elmo Hardy. Pp. 159–242+plates 11–16. 30*s*. The John Murray Expedition, 1933–34. Scientific Reports. Vol. 10, No. 5: Sponges. By Dr. Maurice Burton. Pp. 151–281. 40*s*. Vol. 10, No. 6: Report on the Brachiopoda of the John Murray Expedition. By Dr. Helen M. Muir-Wood. Pp. 283–317+5 plates. 20*s*. (London British Museum (Natural History), 1959.) [312

Other Countries

Optical Society of America, Washington. *Optics and Spectroscopy*, Vol. 6, No. 1. Translation of the Russian journal *Optika i Spektroscopiya*. Pp. iv+84. Published monthly. Subscription rate: 25 dollars plus 3 dollars for postage. (This rate includes the Journal of the Optical Society of America.) (New York. American Institute of Physics, 335 East 45th Street, 1959.) [3011

University of Natal. Wattle Research Institute (South African Wattle Growers' Union—University of Natal—Union Department of Forestry.) Report for 1958–1959 (Twelfth Year). Pp. 51+3 plates (Pietermaritzburg. University of Natal—Wattle Research Institute, 1959.) [3011

Sitzungsberichte der Physikalisch-medizinischen Sozietat zu Erlangen, 79 Band, 1958. Herausgegeben im Auftrag der Sozietät von Hans-Jurgen Stammer. Pp. xviii+210. (Erlangen. Physikalisch-medizinische Sozietat, 1959.) [3011

Institut des Parcs Nationaux du Congo Belge. Exploration du Parc National de l'Upemba. Mission G. F. de Witte en collaboration avec W. Adam, A. Janssens, L. Van Meel et R. Verheyen (1948–1949). Fascicule 56: Amphibians. By Karl P. Schmidt and Robert F. Inger. Pp. 264+9 plates. (Bruxelles. Institut des Parcs Nationaux du Congo Belge, 1959.) [3011

Goteborgs Kungl. Vetenskaps-och Vitterhets-Samhälle. Report of the Swedish Deep-Sea Expedition, 1947–1948. Edited by Hans Pettersson. Vol. 6: Sediment Cores from the West Pacific. Pp. 99–160+14 plates. Sw. kr. 25. Vol. 10: Special Investigations. No. 4: On the Stratigraphy of some Bottom Sections from the Central Pacific. By F. Brotzen and A. Dinesen. Pp. 45–55. Sw. kr. 5. (Goteborg. Elanders Boktryckeri Aktiebolag, 1959.) [3011

Carnegie Institution of Washington. Publication No. 617. Miocene Floras of the Columbia Plateau. Part 1: Composition and Interpretation. By Ralph W. Chaney. Part 2. Systematic Considerations. By Ralph W. Chaney and Daniel I. Axelrod. Pp. viii+237+44 plates. (Washington, D.C.: Carnegie Institution of Washington, 1959.) 4.25 dollars; cloth bound 4.75 dollars. [3011

International Atomic Energy Agency, Vienna. Assistance Through Exchange of Nuclear Scientists. Pp. 8. A Chronological List of Atomic Energy Conferences, Meetings, Training Courses (Issue No. 5). Pp. 22. (Vienna. International Atomic Energy Agency, 1959.) [312

World Health Organization. Bulletin. Vol. 21, No. 1: Epidemiological Basis of Tuberculosis Eradication in Denmark. A Concurrent Comparison of Home and Sanatorium Treatment of Pulmonary Tuberculosis in South India. Pp. v+144. (Geneva: World Health Organization; London: H.M. Stationery Office, 1959.) 6 Swiss francs; 10*s*.; 2 dollars. [312

World Health Organization. Technical Report Series, No. 181 Immunological and Hæmatological Surveys—Report of a Study Group. Pp. 35. 1 Swiss franc; 1*s*. 9*d*.; 0.30 dollars. Communicable Diseases in Schools: a Survey of Existing Legislation. Pp. 65. 2 Swiss francs, 3*s*. 6*d*.; 0.70 dollars. [312

Canada: Department of Mines and Technical Surveys. Geological Survey of Canada. Economic Geology Series No. 17. Tungsten Deposits of Canada. By H. W. Little. Pp. viii+251. 1.50 dollars. Memoir No. 288: Surficial Geology of the Lindsay-Peterborough Area, Ontario, Victoria, Peterborough, Durham, and Northumberland Counties, Ontario. By C. P. Gravenor. Pp. 60+7 plates. 1 dollar. (Ottawa: Queen's Printer, 1959.) [312

United States Department of the Interior: Geological Survey. Water-Supply Paper 1320-E Summary of Floods in the United States during 1953. Prepared under the direction of J. V. B. Wells. Pp. v+341–364. 20 cents. Water-Supply Paper 1505: Surface Water Supply of the United States 1957. Part 3-A: Ohio River Basin except Cumberland and Tennessee River Basins. Prepared under the direction of J. V. B. Wells. Pp. xii+553. 1.75 dollars. Professional Paper 315-A: Geology of Part of the Horseshoe Atoll in Scurry and Kent Counties, Texas. By Philip T. Stafford. Pp. iii+20+8 plates. (Washington, D.C. Government Printing Office, 1959.) [312

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'TEEN-AGE' EDUCATION IN ENGLAND

IN March 1956 the Minister of Education asked the Central Advisory Council for Education (England) to advise him on the education of boys and girls between the ages of fifteen and eighteen. Under its chairman, Sir Geoffrey Crowther, the thirty distinguished members of the Committee have produced a report* which is worthy to take its place alongside such well-known predecessors as Hadow, Spens, Fleming and Norwood. It provides an all-embracing survey of the educational services available to the 'teen-age' population of England and Wales, as well as a penetrating analysis of the very considerable deficiencies in those services.

The Report has clearly been drafted by more than one hand, but by far the larger portion of it is written in the simple prose which would bring commendation from a Churchill or a Gowers. It is a report which, apart from the great importance of its subject-matter, asks to be read because of the attractive way in which it is presented.

Although the main recommendations are likely to stir up discussion which may at times become acrimonious, even the most robust critics of the Report would wish to endorse the Minister's gratitude to Sir Geoffrey and his colleagues for a Herculean task well done.

The Report is concerned with four sectors of educational provision for boys and girls between the ages of fifteen and eighteen—it is not concerned with the youth service, which is being examined separately by the Albemarle Committee—but will, almost certainly, become widely known for two main recommendations. These are the proposals to extend the age at which all children must remain at school—the age would be sixteen and the operative year would be between 1966–67 and 1968–69; and the introduction of county colleges according to a phased programme which would be completed within 15–20 years from now. There are, however, a number of other recommendations which could vitally affect the quality of education provided for teen-agers and which deserve close study. Few will quarrel with them, and in time nearly all should be implemented. Among them are the place of examinations in secondary modern schools, provision for those who leave full-time education at the age of sixteen and who require continued education with a vocational bias through day-release or sandwich courses, the way to combat specialization in school sixth forms (which the Council is prepared to support provided it does not mean over-specialization), the kind of teachers who will be required for the county colleges and the way to find and develop them. The cost of the required reforms would put no unbearable strain upon the Exchequer.

The decision to recommend that the school-leaving age should be raised in the late 1960's was based upon careful analysis of the place of education in a changing world, particularly that relating to secondary education. Whereas in 1894 only about 4 per cent of all boys and girls aged fourteen or fifteen were in school, and only 1 per cent of those aged sixteen and seventeen, to-day about two-fifths of all boys and girls aged 15–17 have education either full-time or for one day a week. Of the 2½ million children in maintained secondary schools, however, only 320,000 of them are aged fifteen or more; the Committee is concerned that the rest of the two million who could benefit should be given opportunities in full-time education as quickly as possible. In 1894 such a proposal would have been regarded as disastrous by parents whose children were regarded as winners of the extra bread which made life tolerable for the family. To-day, few families cannot support their children throughout a longer school education, a situation brought about by marked changes in economic social life of Britain. Compared with 1938, the community is about one-third richer in material wealth. The larger part of the rise in the national standard of consumption has been in the lower income-groups, a change reflected in the physique and health of school-children during the past twenty years. Other influences which make it easier to support a longer school life for children are earlier marriage, earlier child-bearing within marriage, smaller families, longer expectation of life and more opportunities of paid employment for married women.

The position of girls in the age-group studied has probably changed even more than that of boys. Child-bearing and child-care now represent a break in employment for married women rather than an end to it. This has important consequences for teaching as well as other careers. It points to the need for an education which encourages girls to qualify before marriage for careers and to develop interests which they can resume in later life. The early age of marriage points to a radically different concept of how girls of seventeen and eighteen should be treated and educated; more than 4 per cent of the girls with whom the report is concerned are married women.

The pressure of economic change bears upon school life in a number of ways. The earnings of young people have risen much faster in proportion than those of adults, and those who remain at school have to resist the lure of a high immediate wage; and to reconcile themselves to a lower earning capacity, until they are more than twenty years of age, than that enjoyed by those who leave at fifteen. It is evident, however, that they then catch up and go well ahead. Parallel with the need to stay at school as long as possible to obtain long-term economic advantages, there is growing stress on the need for specialist qualifications and for a good general education to secure entry into an increasing number of occupations. This tendency may be reinforced by the fact that the proportion of young people to the whole population of working age is going to rise again after a long period of decline.

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* Ministry of Education. 15 to 18: A Report of the Central Advisory Council for Education (England). Vol. 1: Report. Pp. xxxi+519. (London: H.M. Stationery Office, 1959.) 12s. 6d. net.

It is within this framework that the Committee sets its pattern for the future of teen-age education in the Britain of the future—for what is done in England is likely to be followed by Wales and, probably, Northern Ireland and Scotland. Education is regarded both as a duty that the State owes to its citizens—as part of the Welfare State—and as a means of increasing the economic efficiency of the whole community, and thus as a form of productive national capital investment. The Council welcomes the rapidly growing tendency for many secondary modern scholars to stay at school voluntarily until sixteen and hopes that, by 1965, about half the total numbers of secondary modern boys and girls will be taking extended courses beyond the age of fifteen. For the pupils for whom the General Certificate of Education examinations at Ordinary Level are unsuitable, the Committee recommends that external examinations should be developed experimentally on a regional or local basis.

With this assessment of the state of education and with these recommendations there will be few dissentients. With the major recommendation, that all children should stay at school until they are at least sixteen years of age, there is likely to be considerable disagreement. The Committee might well argue that it was only seeking to justify a decision of Parliament which is already on the statute book. There are many who would argue that all the experience with the lower streams of secondary modern school children since the Butler Act of 1944 should have led the Committee to recommend either that this section of the Act be repealed or postponed almost indefinitely, until much more pressing reforms to improve both the quality and quantity of British educational services be undertaken. Young people of England and Wales would gain much more if the extra teaching and material resources that will become available over the next decade were used to develop nursery schools, reduce the size of primary school classes, improve the quality of education for those taking City and Guilds and National Certificate courses—which in some local technical colleges is unbelievably bad and explains why many students fail to stay the course—to introduce speedily the much-heralded county colleges, to provide hostels so that university students may begin to live university lives and to develop day-release facilities for adult workers in industry.

The more this Report is studied the more one realizes how false a deduction is drawn from such lofty principles. The case for providing facilities for those two-thirds of secondary modern children who voluntarily wish to stay at school until they are sixteen or more and whose parents are equally keen is well made out. The case for compulsorily keeping those in school for whom the extra year (to fifteen) has already proved a burden to themselves and a source of frustration to those who try to teach them, appears to rest on the belief that 'Daddy-knows-best' and on the over-worked moral principle that what is good for some is good for all. Elsewhere in the Report we are told that the greatest number of juvenile delinquents is drawn from boys and girls who are in their last year of school, and that the desire to learn in many young people has only been awakened by their discovery at work of new ambition or personal inadequacy. All who have been concerned with *C* and *D* stream secondary modern youths at work will know how true this is, and would resist any attempt to inflict another year of misery on these unfortunate young people, for whom the academic approach has proved so inadequate in their development.

Here, as the reaction to the proposed raising of the school-leaving age has shown, they would be supported by all those teachers who teach day by day the scholars from lower streams of modern schools. These teachers frankly admit that they have been unable to find the way to interest and educate these boys and girls, and they believe that the only approach which is likely to be successful is through an integrated training scheme in a progressive firm with day-release facilities providing a clear vocational link with everyday work. It is a matter of regret that no member of the Crowther Committee teaches the lower streams in a modern school or deals with them directly in industry once they have left school. It is not without significance, however, that a strong minority of the Committee is far from happy about raising the school-leaving age for certain scholars who are physically or emotionally maladjusted to school life. They should persuade their colleagues to reconsider this controversial proposal and to take evidence from appropriate teachers in secondary modern schools, from industrial training officers concerned with the less intelligent type of existing school-leavers, and, above all, from the young people themselves not only when they are still at school but also six months after they have left.

Of the Committee's second main proposal—to introduce gradually a system of county colleges some fifteen to twenty years from now—the only disagreement will be the timing. The case for day-release was made out in 1918 and has not been contested. It was given a double blessing in 1944. When an important committee now reports that county colleges should come in time—after experiments in a few areas to gain experience and then by a spread region from region—it reveals either lack of conviction, lack of a sense of urgency, or the lack of moral courage to recommend that such a highly desirable reform is one about which Britain as a whole can no longer afford to procrastinate. To declare that more experience is needed before county colleges are developed nation-wide is scarcely complimentary to Bournville, Langside, Rowntree's, Boot's College or the day-continuation activities which are now so well established under local education authorities like Liverpool and others. To infer that the scheme should wait for fifteen-twenty years is less than kind to the boys and girls of to-day and the next generation. To imply that the country cannot yet afford county colleges is flatly contradicted by the Committee's own declaration that, despite the gain in the nation's wealth, we are proportionately spending no more on education—other than what is being done at the universities and in advanced technology—than we were in 1938. This is a bad reflexion on the use of the nation's resources and the amount we are prepared to spend on the education of our young people. Finally, the assumption that the country will have neither the teachers nor the material resources to introduce county colleges before the late 1970's must be set against all that has happened in the past twenty years. A Welfare State has been born in Britain, with health and social services of which we may be proud. The school-leaving age has been raised. Technical and university education have been transformed. Now with the nation's economic pulse beating strongly and steadily, the Crowther Committee asks the British people to wait twenty years before a generally agreed reform is introduced.

Despite major recommendations which are unlikely to get a smooth passage, this Report should be examined in its entirety. The section on specialization in the sixth forms of schools should be read by every university teacher, as well as headmasters and senior masters in secondary schools. The chapters on institutions and teachers should be made into a tutorial course for many members of local education authorities. Finally, because the Report has been specially prepared with the general public in mind, those concerned with mass communication media could perform a public service by presenting its main arguments and recommendations to as big an audience as possible. The Crowther Report will influence the quality of English education for many years to come.

THE BIRTH OF THE ROYAL SOCIETY

History of the Royal Society

By Thomas Sprat. Edited with Critical Apparatus by Jackson I. Cope and Harold Whitmore Jones. (Washington University Studies.) Pp. xxxii + 456 + 78. (Saint Louis, Missouri: Washington University, 1958.) n.p.

Of recent years American scholarship has made many contributions to the history of science in the seventeenth century, prominent among many universities concerned being Harvard, represented by Bernard Cohen and his school, and Washington, the "University Studies" of which include in their wide range this period. The latest of these "Studies", which appears at a time when the forthcoming celebrations of the tercentenary of the foundation of the Royal Society make it of particular interest, is a re-issue of Thomas Sprat's "History of the Royal Society", furnished by the two editors with an introduction and extensive notes.

Thomas Sprat, born in 1635, graduated at Oxford, where he made some stir as a poet but none as a follower of the newly established pursuit of experimental science. He was, however, taken by Wilkins into the circle of active promoters and partisans of the new learning and was duly elected a Fellow of the newly formed Royal Society in 1663, which was no warranty of scientific prowess, as evidenced by the fact that the poets John Dryden, John Denham and Edmund Waller were admitted at the same time. He had close personal relationships with many of the distinguished founders of the Society and it was their influence, no doubt, that led him to write his "History of the Royal Society", which, published in 1667, is a history of the foundation of the Society and a defence against the scoffers; the misunderstanders, such as Sorbière; and the classical and religious carpers. The full title runs "The History of the Institution, Design and Progress of the Royal Society of London, For the Advancement of Experimental Philosophy". Sprat wrote largely as an apologist; as he says in his "Advertisment to the Reader": "and the Third (part) chiefly contains a Defence and Recommendation of Experimental Knowledge in General: So that it is only the Second Book that peculiarly describes their Undertaking". This "Second Book" which, however, occupies considerably more than half the volume, contains accounts of various scientific concerns of the Society, including, in particular, the "Method For making a History of the Weather", by Robert Hooke.

The book, written in a clear and pleasing style, attracted considerable attention, went into several editions, and was translated into French in 1669. In 1825, de Clugny wrote of it and the Royal Society: "Sprat mérita de devenir membre de cette société, et fut chargé par elle d'écrire son histoire, lorsqu'il devint nécessaire de se concilier l'esprit du public, qui croyait voir dans toute assemblée quelconque des sociétés pour rétablir la suprématie du pape. Son *Histoire de la société royale* . . . triompha de tous les pamphlets qui furent publiés contre elle. Cet ouvrage a été san doute surpassé, quant aux faits, par Birch, et plus recamment par Thomson; cependant il est écrit avec tant de pureté et d'élégance, qu'il est encore recherché". The reference to the supremacy of the Pope is somewhat surprising!

The text in the issue now before us is a facsimile, actual-size reproduction of the text of the first edition of the book, and the "Introduction" discusses, clearly and with ample references, the circumstances which attended the publication of the book and the nature of its contents, as well as the style in which it is written. Among the notes, which bespeak wide reading, those on Stubbe's acrimonious attacks on the Royal Society are of particular interest. In a short appendix the origin and nature of these attacks are further discussed, with references which do not include, however, Harcourt Brown.

In the "Foreword" the editors remark that Thomas Birch's "History of the Royal Society" is 'crippled' for want of an index, a complaint with which every student of seventeenth-century science will wholeheartedly sympathize. In fact, no Ph.D. degree could be more serviceably earned than by the compilation of such an index which, if well done, would demonstrate the necessary scholarship. An important earlier "Washington Study", Richard Foster Jones's "Ancients and Moderns", is likewise robbed of its full value by lack of an index. It is remarkable, in view of their specifically expressed opinion concerning Birch, that the editors should not have provided Sprat with an index. It is to be hoped that, should a re-issue of the book be called for, which is not unlikely, an adequate index will be furnished.

E. N. DA C. ANDRADE

MICROBIAL VARIATION AND RUSSIAN EXPERIMENTS

Microbial Variation

Edited by V. D. Timakov. Translated from the Russian by G. H. Beale. Pp. x+202. (London and New York: Pergamon Press, 1959.) 40s. net.

THE book contains twenty-two papers by workers at the Gamaleia Institute of Epidemiology and Microbiology on the subject of 'directed variation'. The investigations described fall into two main groups: studies of the consequences of growing one bacterial species on killed cultures of a different species in an attempt to explain the formation of new species in bacteria; and examination of the changes brought about in organisms as a result of submitting cultures to the effects of antibiotics, bacteriophages, immune sera, etc., with the object of obtaining avirulent organisms capable of immunizing against pathogenic bacteria. The thesis put forward by Prof. Timakov in the introductory article is that organisms undergo variation under such conditions and that the nature of the variation can be directed by the environmental conditions imposed.

The more controversial material is contained in the first nine papers, concerned with the variation of an organism 'directed' by growth in the presence of dead cells of another species. Sufficient experimental detail is provided to allow some assessment of the significance of the results or to provide the recipes for attempts at reproduction of the experiments. In essence the experiments involve the growth of a large inoculum (10^{10} cells) of, say, *Escherichia coli* in a medium containing a suspension of killed cells (density 10^8–10^9 cells/ml.) of, say, *Salmonella breslau*. Transfers are then made at intervals of seven days (details of times and temperatures are not stated in all papers) and variants appear after 6–15 transfers. Variants isolated after 6–7 transfers are unstable and lose their 'new' properties on cultivation in normal media; variants isolated after the fifteenth transfer are stable and possess new, stable characteristics. Examination of the properties of these stable variants shows that they have acquired some of the characteristics of the killed cells on which they were grown. In the example cited, cells are obtained which are agglutinated by anti-*breslau* but not by anti-*coli* serum, and have become virulent. Other variants are found to have characteristics intermediate between those of *E. coli* and *S. breslau*. In his introduction, Prof. Timakov suggests that this is not the change of one species into another, since the *breslau*-like variant differs from *S. breslau* in being less able to produce infection when given orally; consequently, the experiments are said to demonstrate the production of a new species. It all depends on how you define a species, and there is considerable specious argument on this topic.

Such 'directed variations' are accomplished using various typhoid, paratyphoid, dysentery and coli-form organisms. The editor remarks that "Certain objections which are not without foundation have been raised in connexion with this work . . . some workers believe that the bacteria . . . with characters of *S. breslau* were nothing more than germinated heat-resistant forms of *S. breslau*". One wishes that more details were provided to refute this belief; we are simply told that the suspensions were tested for sterility over a period of from fourteen days to six months. No practical details of bacteriological technique or quantitative data for controls are given. It is possible that something akin to transformation might be taking place in these experiments, and one paper deals with the directed variation of *E. coli* grown in the presence of nucleoproteins from pathogenic bacteria, but the work of Griffiths, Avery, Macleod and McCarty is dismissed in three lines as "inconclusive as proof of the occurrence of inter-specific variation". There is little attempt to explain what might be happening in these experiments, other than to emphasize that the formation of new species can be directed in these various ways.

The book provides an interesting account of the work and views of certain Soviet biologists. Readers' reactions will be largely conditioned by their training and beliefs, but, at least, they now have sufficient data provided for them to attempt to direct variation for themselves.

E. F. GALE

NON-METALLIC BRITTLE MATERIALS

Mechanical Properties of Non-Metallic Brittle Materials

Edited by W. H. Walton. (Proceedings of a Conference on Non-Metallic Brittle Materials organized by the Mining Research Establishment of the National Coal Board in consultation with the Building Research Station (D.S.I.R.), and held in London, April 1958.) Pp. xi+492. (London: Butterworths Scientific Publications; New York: Interscience Publishers, Inc., 1958.) 90s.

THE variety of brittle materials is immense; some of them are heterogeneous even on a macroscopic scale, and some are anisotropic. Both features make their study difficult. Yet a knowledge of the properties of these materials is of great importance to a variety of industrial processes and it was with this idea in mind that the Mining Research Establishment of the National Coal Board and the Building Research Station organized a conference in April 1958. The Proceedings of this Conference present, probably for the first time, a series of papers which demonstrate the immense amount of work which is going on in this field and also emphasize the difficulties of elucidating the properties of brittle materials because of the presence of a large number of varying parameters. The format of the Proceedings in general follows the course of the conference.

At Session 1, "Strength in Compression, Tension, Bending and Shear" was considered and papers were presented giving experimental results on the strength of cubes of coal under uniaxial compression and under triaxial compression, the strength of bricks and a review of the micro- and macro-strength of glass. Other subjects considered were the strain-stress relations and breakage of rocks, of their properties in conditions of triaxial stress and also the porosity and strength of brittle solids with particular reference to gypsum. These papers show a real attempt to correlate experimental results with theory, but it must be appreciated that many of the derived formulæ must, for the present, represent only an empirical interpretation of the properties and that much work is still to be done before the course of changes induced by applied stress can be forecast with any degree of accuracy.

Session 2 was devoted to consideration of "Elasticity and Creep". Papers were given on the elasticity, creep, shrinkage and the effect of rate of loading on some mechanical properties of concrete. The strength of ceramic materials under impact testing and sonic testing, and the stress-strain relationships in heavy clay products were discussed, while the factors effecting the thermal shock resistance of high-temperature materials were also considered. Measurements of the rheological behaviour of coal, together with the measurements of its dynamic mechanical properties, were compared with the paper presenting the elastic and plastic properties of carbon and graphite.

In the third session the conference went on to consider "Dynamic Loading, Impact and Fragmentation". Here papers were presented on the exploratory tests on the strength of concrete beams under pulse loads, impact testing and roller bit penetration. The breakage of natural materials was considered generally and in detail with the fragmentation of explosive detonated in stone blocks and with the partition of energy in a blasting shot. From the coal industry, studies of the size distribution and relationship between dust formation and coal strength provided an alternative approach.

Session 4 was devoted to a consideration of the "Action of Tools". There was a basic paper on friction between coal and metal surfaces and the forces required to penetrate the brittle material with a wedge-shaped tool. This was followed by a paper on the effect of lateral pressure on the cutting of coal by wedge-shaped tools and a consideration of both the theoretical and practical aspects of coal ploughing.

The discussions which followed papers and sessions have been admirably condensed and the whole volume will be an important work on the shelf of all who are concerned with understanding the processes in which the properties of brittle materials have to be taken into account. DAVID TRAIN

CONSERVATION IN AMERICA AND AFRICA

Conservation of Natural Resources
Edited by Guy-Harold Smith. Second edition
Pp. xi+474. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1958.) 68*s*. net.

The Conservation of Natural Resources
By Richard C. Haw. Pp. 256+36 photographs. (London: Faber and Faber, Ltd., 1959.) 30*s*. net.

IN every part of the world, though in varying degrees, the now obviously rapid increase in population consequent upon the fact that the spread of the knowledge and practice of death control has outstripped knowledge and practice of birth control is forcing attention on the need to conserve or to use wisely all natural resources. In the second edition of "Conservation of Natural Resources" the editor, Guy-Harold Smith, repeats the warning, "every individual company, or organization must share in the task of preserving the resource base upon which the American economy has been built. What is needed is an enthusiasm and an understanding of the essential nature of resources and the measures necessary to save from wasteful destruction our national heritage." It may be claimed that conservation in America was a by-product of the great economic depression of the nineteen-thirties which happened to coincide with disastrous seasons producing dust-bowl conditions over vast areas. Although ghost-towns already spoke eloquently of the exhaustion of mineral riches, abandoned farms of the retreat from the 'frontier' or 'pioneer fringe', conservation became identified with the prevention of soil erosion. An America which had been taught to think in square-mile blocks had to realize that Nature abhors a straight line, and to find answers in contour ploughing, strip cropping, restitution of grass cover thoughtlessly ploughed or woodland unwisely cleared. Now conservation has come to mean much more. The sixteen authors of this survey cover in its eight parts the land (with soil, irrigation, and grassland), the forests, water, minerals, wild life, recreational resources (linked with the conservation of man) and so to national, state and local planning.

Prof. S. S. Visher has an interesting introductory chapter on the public domain. It is perhaps not commonly recognized that practically all United States territory, except for the closely settled land of the original thirteen States, has, at one time, been Federal property. Forty-one per cent was sold to individuals for cash, 27 per cent granted to homesteaders, 13 per cent given to the railroad companies, 12 per cent given to education authorities, 1 per cent for canals, leaving 6 per cent, mainly of wet lands, which passed into ownership of the States. In 1956 more than 400,000,000 acres remained or had reverted to public ownership, being a total not far short of that in improved farm land. Thus Federal and State authorities are able to give a lead to conservation in the national forests and the great grazing lands as well as in national parks and Indian lands. But on privately owned land there are conflicting interests. Individual success is still measured largely by wealth: the obvious temptation is the quick fortune from 'mining', whether it be of minerals, the soil, or the forests. Technological advances will take care of the future and with the present over-production of agricultural commodities, those who preach conservation or teach it in the universities have by no means a pre-converted audience. This fine book should serve them well.

Although with an identical title, Mr. Richard C. Haw's book is very different. He is an instructor in the Domboshawa Government School at Salisbury in Southern Rhodesia: he writes essentially of the African veld and of the vital, urgent, necessity of its proper management if Africa is to survive. He uses the University Farm at Salisbury West as an example of planned layout; chapters which follow are practical guides to surveying, soil conservation, manuring, reclamation, road construction and water conservation. It is significant that he, too, does not forget the importance of wild life. Since so much of the world's problem lands lies in the seasonally dry sub-tropics, Haw's book deserves a wide public in other continents. L. DUDLEY STAMP

THE ARMAMENT RESEARCH AND DEVELOPMENT ESTABLISHMENT

OPEN Days were held for the first time, during November 3–6, at the Armament Research and Development Establishment, Fort Halstead, Sevenoaks, Kent. This Establishment was part of the former Ministry of Supply and from November 23 has been part of the War Office. The recent changes in organization made in Government departments prevented the attendance of a Minister of the Crown, and the official welcome to visitors to the Open Days was extended by Lieutenant-General Sir John Cowley, controller of munitions.

The Open Days were planned with the object of inviting as guests a wide representative selection of people who had not previously had the opportunity of visiting the Establishment. Although security restrictions inevitably affected the scope of the exhibition, an excellent impression of the wide variety of work undertaken by the Establishment was afforded to visitors.

More than three thousand visitors attended the exhibition during the four days. During November 4–5 the visitors included Government officials and Service officers, representatives of Commonwealth countries, attachés at foreign embassies, members of local authorities, teachers at universities, technical colleges and schools, and representatives of industry. On November 6, the Establishment was open to families and friends of the staff. Previously, on November 3, the Establishment welcomed about fifty representatives of the national, local and technical Press and of sound and television broadcasting services. It was evident that the exhibition attracted very considerable interest from the general public.

The Armament Research and Development Establishment was created on January 1, 1955, by the amalgamation of the Armament Research Establishment and the Armament Design Establishment. The director of the Establishment is Dr. D. H. Black. The work of the first-named Establishment was surveyed in a previous article in *Nature*[1]. Up to the end of the Second World War, the history of the Armament Research and Development Establishment and its predecessors is essentially the story of the development of high-explosive munitions. The post-war years have brought considerable changes in the work undertaken. Under the direction of Sir William Penney, the development of Britain's first atomic bomb was undertaken within the Armament Research Establishment organization at Fort Halstead. This project led to the formation of the Atomic Weapons Research Establishment at Aldermaston, now a part of the United Kingdom Atomic Energy Authority. The part played by conventional weapons has been greatly affected by the development of nuclear explosives and weapons. The Establishment continues to undertake highly important work on conventional explosives and weapons, but has also extended its responsibilities to newer branches in the armament field.

The Establishment undertakes research, and designs and develops weapons and ammunition for all three Armed Services. Equipment for which it is responsible is extremely diverse; this includes guns together with their mountings and ammunition, fuses, small arms and their ammunition, mines and pyrotechnics. The Establishment is also concerned with work on guided weapons. Much basic research, both theoretical and experimental, is pursued. This research is not always immediately related to particular weapons, but often leads later to applied research aimed at the design, development or improvement of specific weapons. Basic and applied research are undertaken in a wide variety of fields; these include high explosives, ballistics, metallurgy, radiography, electronics, electronic computers, and the mechanics of fluids and solids. Much of the research done falls within the ambit of the physical sciences, and applications outside the armament field often occur.

More than a hundred individual items were listed in the guide to the exhibition, and these ranged from topics in basic research to actual demonstrations of weapons. It is not possible to give here an overall description. Instead, very brief accounts will be given of some current researches in a few important fields of general scientific interest.

Hypersonic Aerodynamic Flow

Laboratory investigations of the hypersonic flow of air past a body are very much more difficult than those of supersonic flow. Not only is it difficult to achieve the true stagnation temperatures at the correct Mach and Reynolds numbers of the flow, but also the entire physical picture is now considerably complicated by the occurrence of so-called real gas effects, such as dissociation, ionization and relaxation. The transition from the supersonic to the hypersonic régime occurs at a Mach number of about 5, but all the characteristic features of the latter régime may only become well developed at a much higher Mach number of, say, 15. The development at the Establishment of the light-gas gun-tunnel is an attempt to obtain a satisfactory laboratory facility for use in these investigations. A light gas at high pressure drives a light piston down a sealed gun barrel, and thereby gas (typically air) in front of the piston is compressed and is shock-heated. On completion of sufficient compression in the driven gas, this gas (now hot) is automatically released through a throat, to give expanded flow at Mach numbers between 8 and 15 in the working-section of the tunnel. The running-time in the working-section is about 50 ms. Schlieren photographs are taken, and total force, pressure and heat-transfer measurements are made of the gas-flow past models.

A low-density continuous-flow wind-tunnel with a plasma heater is in operation at the Establishment and is shown in Fig. 1. This facility caters for the simulation of conditions for aerodynamic flow at Mach numbers up to 12 and at altitudes up to 50 miles. Another facility is a small shock-tube in which shocks, travelling initially at Mach numbers of the order of 150, are set up by magneto–gas dynamic impulses.

Thermal Properties of High Explosives

Aerodynamic heating in high-speed flight may result in explosive components acquiring tempera-

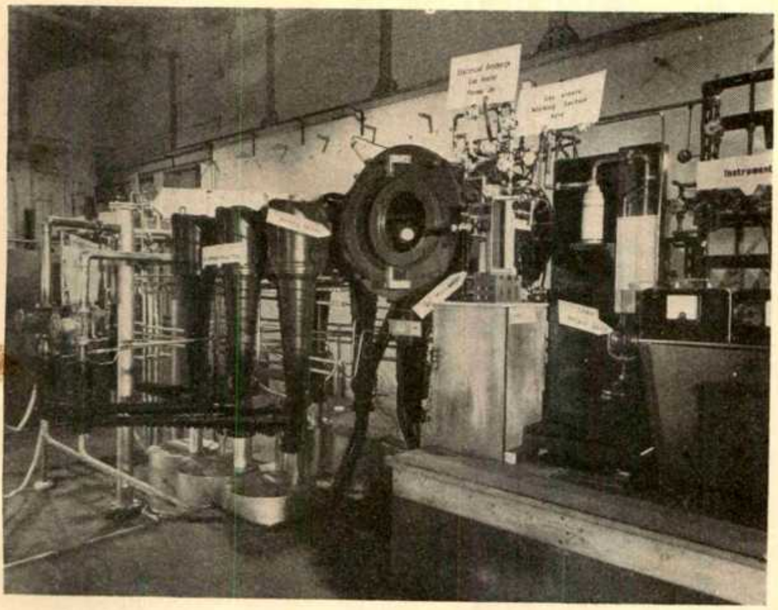

Fig. 1. Low-density continuous-flow wind-tunnel

tures above normal. In order to examine the functioning and safety of these components under these conditions, the physical and chemical properties of high explosives are being investigated at elevated temperatures. These investigations include studies of thermal decomposition, thermal conductivity and sensitiveness.

Thermal decomposition of high explosives is investigated by the measurement of the rate of evolution of gas. This technique has been used particularly for RDX (cyclo-trimethylene trinitramine) and TNT mixtures. The main reaction is the first-order decomposition of RDX in solution in TNT. When RDX is suspended in a saturated solution of RDX in TNT, however, an additional reaction occurs in which it is believed that the TNT decomposes on the surface of the suspended RDX. The heat evolved during the decomposition of high explosives is determined by the rise in temperature of a copper canister, containing the explosive, placed in an electrical furnace the temperature of which is high enough to cause decomposition. In the case of tetryl (2 : 4 : 6-trinitrophenyl methyl nitramine) and TNT, the heat of decomposition is evolved in two stages, indicating the existence of a stable intermediate.

Thermal conductivity is determined by means of an apparatus in which use is made of Callendar's method. The examination of RDX/TNT 60/40 below the melting point of TNT shows that the thermal conductivity decreases with temperature rise.

Impact sensitiveness of a high explosive shows no variation with temperature if this is below its melting point. Above the melting point, sensitiveness depends upon the confinement of the sample. This is illustrated by the results obtained from tests made on small samples of molten high explosive placed upon an anvil. On impact, if the sample is ejected from the surface of the anvil, then the apparent sensitiveness is decreased, as compared with that of the solid high explosive ; but when the sample is confined within a recess in the surface of the anvil, the reverse is the case.

Metallurgy

In the field of ferrous metallurgy, very high strength Si-Cu-Mo-V steels have been developed by the application of research into the effects of various alloying elements on volume changes at transformation temperatures, on martensitic breakdown during tempering, and on hardenability. These steels have tensile strengths greater than 140 tons/in.^{2}, good ductility and impact resistance, and are very resistant to tempering. The suitability of these steels for various potential applications is being evaluated ; these applications include vehicle springs and torsion bars, aircraft high-tensile steel forgings and castings, missile components fabricated from sheet, and cold extrusion tools.

Research on materials at high temperatures has enabled considerable progress to be made in producing high-quality arc-melted molybdenum alloys and in working these alloys to bar, rod and sheet by means of forging, swaging, extrusion and rolling processes. The ductile–brittle impact transition temperature has been lowered to 225° C. by a metal-working process, but by using electron-bombardment melting for zone-refining, this temperature has been further substantially lowered to −140° C. in purified single crystals and to −80° C. in purified polycrystals. Progress is also being made in the protection of molybdenum at high temperatures, and promising results are being obtained with multi-layers of electro-deposited coatings in combination with ceramic.

The occurrence of repeated high stresses in certain engineering structures has necessitated design for a limited life. In this connexion, a push–pull machine has been developed for high-strain fatigue testing that is capable of producing failures in specimens down to within a single cycle.

Plasma-jet and transferred arc torches have been developed, and their use for the spraying of high melting-point metals as well as ceramics and other refractory materials is receiving considerable attention.

Ultra-high Speed Radiography

In certain armament investigations, the presence of high-luminosity, smoke and flying debris precludes the use of high-speed photographic techniques for observing events. Radiographic techniques must then be used instead. Techniques are now available for taking single-shot radiographs through approximately 2 in. of aluminium, with exposure times of less than 1 μsec. Some current research is concerned with the development of megavoltage systems which make it possible to penetrate 3 in. of steel, with the same exposure time. These techniques have proved useful in a wide range of explosion studies and have also been used in studies of industrial processes such as the flow of molten steel into moulds of complicated shape.

Other work involves the use of the Establishment's 5-MeV. linear-accelerator X-ray generator. This

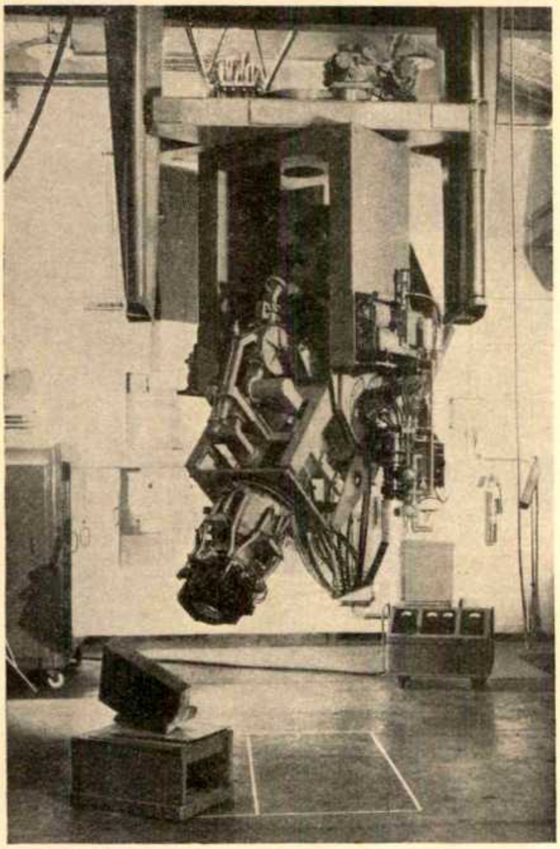

Fig. 2. 5-MeV. linear electron accelerator for radiography. (Penetration, 14 in. of steel; output, 600 röntgens/min. at 1 m.)

equipment (Fig. 2), although primarily used for the radiographic examination of heavy sections (up to 14 in. of steel, with a wire-sensitivity of 0·5 per cent), has also been used in strobo-radiographic work. The generator is operated with a pulse of 1·8 μsec. duration at a maximum frequency of 500 c./s. In this way, studies have been made possible of the valve and cam movements in a single-cylinder internal combustion engine operating at 1,800 r.p.m. Another application made of the technique is to the study of the response of enclosed systems to vibration.

Mechanics of Solids

The propagation of stress waves in solids and the mechanical properties of materials under dynamic conditions are subjects of fundamental importance in connexion with the investigation of certain problems of explosion or impact loading. Theoretical and experimental research is being done in this subject for various materials, including metals and visco-elastic solids such as brittle plastics and other high polymers. Of particular interest is the application of modifications of the Hopkinson pressure-bar technique to compression tests of softer metals and visco-elastic solids. In this type of mechanical test, a short cylindrical specimen is inserted in a 'split' pressure-bar. At one end of the pressure-bar, a stress pulse is generated by the detonation of a small explosive charge. Measurement of the change in time of the displacement produced at the other end of the pressure-bar enables stress–strain characteristics of the specimen to be derived for durations of applied loading of 10–50 μsec. The test has proved particularly useful in the study of visco-elastic solids. The dynamic value of Young's modulus that is derived in this way is in good agreement with values obtained from vibration and stress-wave propagation studies. For the softer metals, it is found that whereas the initial flow stress is significantly greater than that observed at low strain-rates, further deformation at these ultra-high strain-rates appears to be accompanied by unsuspected effects which are being further investigated. Considerable plastic flow in these materials occurs during the fall in stress, and in some cases at sufficiently large strain the dynamic stress–strain curve appears to fall below the static one.

Computational Techniques

A Ferranti Mark I* high-speed electronic digital computer, *Amos*, is used extensively to obtain numerical solutions to mathematical problems arising in the course of studies in a wide variety of scientific and engineering fields. Some examples are the mechanics of fluids and solids and the analysis of individual (and also systems of) weapons.

One particularly interesting recent development is a new computational technique for the numerical integration of systems of non-linear wave-propagation equations in three independent variables. The integration is made along bi-characteristic curves of the equations. Partial differential equations of this (or equivalent) type are of common occurrence in mathematical-physical investigations and methods of solving them are therefore extremely important. Examples occur in the theory of supersonic flow of fluids and also in theories of plastic flow in metals and other solids. So far, the technique has been applied only to the investigation of the steady supersonic aerodynamic flow around a body with the shape of a delta-wing. The theoretical predictions of pressure distributions agree very well with experimental results obtained in wind-tunnel tests. In principle, the technique could be extended to include equations of similar type but in four independent variables, but the computational work would then be severely increased.

Amos has also been used to make important advances in the design of lens optical systems. The important optical aberrations cannot all be reduced to satisfactory minima if the system contains only a few elements, and the consequent need for complex systems introduces difficult problems of design. The recent development of a new and efficient technique of optimization has made possible the reduction of these aberrations to several orders below that previously achieved.

In each of the above investigations a new analytical approach has provided the basis of the computational technique, but it is interesting to note that the use of the technique was possible only with the aid of the computer.

For certain types of numerical problems formerly solved on desk computing machines, it has been shown that the use of the full code of general purpose computers may often be advantageously avoided, particularly with less experienced programmers in mind. A simplified code, 'Intercode', has been developed for use on *Amos*, and several highly successful applications have been made of this procedure.

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[1] *Nature*, 172, 1075 (1953).

GENERATION OF TEMPERATURES BELOW 1° K.

On December 11, 1959, the Physical Society Low-Temperature Group held a symposium at the Institute of Physics on "Generation of Temperatures Below 1° K.", with Dr. P. H. Sykes as chairman. As on previous occasions, the programme was arranged to give plenty of time for questions and discussion. Very little unpublished work was presented, the purpose being chiefly to give an account of established principles and techniques to members of the Low-Temperature Group interested in this field. A detailed report of the proceedings is to be given elsewhere; the following is a brief outline of the material presented.

Following the chairman's introductory remarks, Mr. G. Davey (Clarendon Laboratory, Oxford) discussed the use of liquid helium-4 as a constant-temperature bath down to about 0·74° K., which is effectively the lower limit since the vapour pressure is very small there and is decreasing rapidly with temperature. If only a small pumping speed is to be used, the evaporation rate and hence heat influx to the bath must be drastically reduced, which entails the use of a narrow constriction in the pumping tube just above the bath, to avoid excessive heat influx through the superfluid film. Alternatively, to avoid a constriction, and allow a larger heat influx, a very large pumping speed must be used. Mr. Davey described an apparatus based on this principle, which used a pumping speed of 900 l./sec. and reached a temperature of 0·74° K. For comparison, a temperature of 0·75° K. can be obtained with a speed of 10 l./sec., and a suitably small constriction. The choice of method depends on the type of experiment and the equipment available.

The lower limit of constant-temperature baths can be extended to nearly 0·3° K. by use of the light isotope helium-3, owing to its much higher vapour pressure. Mr. M. Hobden (lately of the Clarendon Laboratory, now at the Royal Radar Establishment, Malvern) summarized the properties of ^{3}He relevant to this purpose, and described several ^{3}He cryostats, which, because of the scarcity of the substance, have come into use only in the past few years. In the 'single-shot' type, ^{3}He is first condensed into the cryostat and the vapour then pumped off to reduce the temperature. An alternative arrangement is the circulating type, wherein the gas is condensed in a vessel immersed in a helium-4 bath, and the liquid allowed to drip (through a needle valve or other high resistance) into the experimental space, where it evaporates and is pumped off, then compressed and re-condensed to continue the cycle. The latter method is more complicated but needs less ^{3}He, and can deal with a larger heat influx. There are no difficulties associated with a superfluid film in ^{3}He, but it must be carefully purified (notably of tritium and ^{4}He) and needs a fairly elaborate gas-handling system. On the whole, the cryostats are simple and convenient, and will undoubtedly come into increasing use.

Dr. E. Mendoza (University of Manchester) gave an outline of the principles and techniques of the adiabatic demagnetization method, whereby paramagnetic salts may be cooled to about 10^{-3} ° K., and other substances, through them, to about 10^{-2} ° K. The entropy of a paramagnetic salt may be reduced by applying a magnetic field, or by decreasing the temperature. The basic principle is therefore to magnetize the salt isothermally, lowering its entropy, and then to remove the magnetizing field isentropically, when its temperature must drop. The final temperature is proportional, ideally, to the ratio of the initial temperature to magnetizing field, typical starting conditions being 1° K. and 10^4 gauss. In practice, the final temperature is limited by the interactions between the ions, which may be reduced by using a magnetically dilute salt. Temperatures are measured by various methods, most commonly by determining the susceptibility of the salt, which is related to the temperature in the ideal case through Curie's Law. The absolute-temperature scale is established by experiments involving Carnot cycles. The cold salt will warm up at a rate determined by its specific heat and the heat influx, and one of the difficulties is frequently found to be the reduction of unwanted heat influxes due, for example, to mechanical vibrations or to desorption of exchange gas as the salt warms up. Dr. Mendoza also discussed means of obtaining isothermal conditions in the demagnetization region, for example, by isothermal demagnetization or by use of the magnetic refrigerator. Another problem encountered is the design of 'heat switches' to make and break thermal contact, among which are the superconducting, 'bath plug', and ^{3}He rectifier types. In the discussion, Dr. K. Mendelssohn (Clarendon Laboratory) mentioned the use of adiabatic magnetization of a superconductor for obtaining temperatures of a few tenths of a degree.

The more esoteric technique of nuclear adiabatic demagnetization was described by Dr. N. Kurti (Clarendon Laboratory). Since the limiting temperature in the demagnetization method is determined by the magnetic interactions present, much lower temperatures can be reached by reducing these, namely, by choosing a substance the entropy of which at low temperatures is determined not by the electron spins but by the nuclear spins, which have magnetic moments smaller by a factor of about 10^3. To obtain the same entropy reduction as in the electron spin case, it is necessary to use a ratio of magnetic field to initial temperature about 10^3 times larger. In one experiment, for example, a specimen of copper was demagnetized from 0·012° K. with a field of 30 k. gauss, the low starting temperature being achieved by 'ordinary' demagnetization of chrome potassium alum. Dr. Kurti explained some of the problems of technique, and of interpretation of results, emphasizing that the lowest temperature reached, about 10^{-6} ° K., was that of the nuclear-spin system, not of the lattice or the conduction electrons. To obtain cooling of the bulk material, it would be necessary to insert a heat switch between the electronic and nuclear stages. Even without this, it should be possible to cool, and do experiments on, the conduction electrons down to at least 10^{-3} ° K

Dr. H. London (Atomic Energy Research Establishment, Harwell), in a paper entitled "More Abstruse Techniques", analysed the possibility of using the reversible adiabatic dilution of liquid ^{3}He with liquid ^{4}He. Such a dilution, in analogy with the adiabatic expansion of a perfect gas, leads to a cooling which is enhanced by the latent heat of mixing. A complete analysis of the method is not simple, and the possibilities may be limited by the presence of the phase separation region in liquid ^{3}He–^{4}He mixtures. In practice, reversible dilution can be achieved by introducing superfluid ^{4}He through a superleak into pure liquid ^{3}He, or into a ^{3}He–^{4}He mixture. One might expect to work with temperatures around 0·1° K.—perhaps even lower—although no direct experiments have been carried out at this stage. An advantage of the method would be that it does not require the use of magnetic fields, on the other hand, if it were to be used as a continuous refrigerator the system would be quite a complicated one.

The symposium ended with a description by Dr. D. F. Brewer (Clarendon Laboratory) of two recent sets of experiments on helium-3 carried out at the Ohio State University, using the adiabatic demagnetization method for cooling. Down to 0·085° K. the specific heat of the liquid was found to have no anomaly such as occurs with liquid helium-4, and appears to vary linearly with temperature below about 0·1° K. In this respect the liquid behaves like a Fermi–Dirac gas with effective mass twice that of a ^{3}He atom. In other respects, such as its increase of specific heat with pressure below 0·16° K. and its negative expansion coefficient, it is unlike a perfect Fermi-Dirac gas. These results can be explained theoretically. Experiments on the melting curve confirm by direct measurement the presence of a minimum at 0·32° K. which had already been inferred from other experimental and theoretical sources. The region of the melting curve below the minimum is inaccessible to measurement by the usual methods, and hence evaded observation for some time; the measurements described were carried out by use of a strain gauge as pressure transducer, which circumvents this difficulty. They confirm the existence in solid helium-3 at low temperatures of forces additional to the magnetic spin-spin interaction.

D. F. Brewer

FOOD RESTRICTION AND SALT PREFERENCE IN RATS

By HANS KAUNITZ, LESTER M. GELLER, CHARLES A. SLANETZ and RUTH E. JOHNSON

Department of Pathology, Institute of Comparative Medicine, and the Division of Neuropathology, College of Physicians and Surgeons, Columbia University, New York

MOST human beings, as well as many other animals, will consume an appreciably greater amount of salt, if it is available, than is required for survival, growth or reproduction. The possible relationship between heavy salt intake and hypertension has been recently re-emphasized[1], while the beneficial effects of high salt intake in such conditions as profuse sweating[2], and in adrenal[3] and certain cases of renal[4] insufficiency are well known. It has been suggested that the salutary action of salt in these cases may not be due to a simple replacement phenomenon, but rather to the consequences of a stimulating action of sodium chloride upon certain body tissues[5]. Presumably such an enhancing effect of salt might also influence certain behavioural responses of the organism to its environment.

The preference of rats for solutions of sodium chloride, particularly in isotonic concentration, as against water, has been well established[6–9]. Our interest in this area lay in investigating possible differences in the level of fluid intake of rats drinking salt solution, as compared with fresh-water drinkers, under conditions of marked and moderate food restriction, and also in studying the possible effects of excessive salt intake upon survival, growth (body-weight), reproduction, and weights of certain organs. Studies of the speed of eating and maze activity of drinkers of salt solution will be reported later.

Male rats from our stock colony (Columbia–Sherman strain, arbitrarily bred as a closed colony for at least fifteen generations) were maintained from weaning (24 days of age) on a purified diet[10] consisting of 30 per cent alcohol-washed casein, 20 per cent lard, 44 per cent dextrose, 2 per cent cellulose, 0·5 per cent calcium carbonate, and liberal amounts of all known vitamins. To this diet was added 4 per cent of U.S. Pharmacopeia salt mixture XIII, containing enough sodium chloride to allow a level of approximately 0·15 per cent of the last named in the final diet. This quantity of salt had been found to be adequate for permitting maximal growth, survival and reproduction in our rat colony, and it was our intention in these experiments to avoid any limiting factors in the diet *per se* which might interfere with such functions.

Each animal was housed in a roomy cage, on wood shavings. The cages were supplied with water bottles bearing machined stems which minimized dripping sufficiently to permit reliable determinations of liquid intake to be made by weighing the bottles to an accuracy of ± 1 gm. All young were earmarked. and then weighed at 28, and again at 35, days of age. Two matching groups of forty rats each were formed having identical average weights at 28 and again at 35 days of age, a procedure which strongly implied similar average growth-potentials in both groups. One group continued to drink water, and the other was offered a bottle of 1 per cent salt solution and one of water.

Shortly thereafter an attempt was made to equate the groups with respect to temperamental or personality characteristics. Each rat was placed in a Lashley III maze (eight *culs de sac*)[11], and observed

Table 1. AVERAGE DAILY FLUID INTAKE OF RATS DRINKING 1 PER CENT SALT SOLUTION AND WATER, OR WATER ALONE, UNDER DIFFERENT CONDITIONS OF AVAILABILITY OF FOOD

Group	No. of rats	Age* (days)	Weight* (gm.)	Average daily fluid intake (c.c)								
				Free access to food (14 days)			Weight reduction (7 days)			Maintenance of reduced weight (21 days)		
				Water	1 per cent salt	Total	Water	1 per cent salt	Total	Water	1 per cent salt	Total
Salt solution and water	20	282	471 ± 18†	7 ± 1·2	34 ± 3·6	41 ± 3·1	6 ± 0·8	59 ± 5·7	65 ± 5·5	5 ± 0·8	65 ± 5·5	70 ± 5·7
Water only	20	282	459 ± 21	25 ± 1·2	—	25 ± 1·2	23 ± 2·3	—	23 ± 2·3	25 ± 1·6	—	25 ± 1·6

* Age and mean weight of animals at beginning of fluid intake determinations (day 1 of period of free access to food)
† Standard error of the mean.

for 5 min. on each of four consecutive days. As possible indicators of the nature of the response of each animal to a novel situation, the time required to leave the starting box, the number of alleys investigated, and the distance traversed toward the goal box, as well as the number and consistency of boluses dropped—an 'emotional' indicator to some workers[11]—were recorded. From the originally matched groups of forty rats each, two sub-groups of twenty were selected on the basis of the similarity of their behaviour in the maze. The final groups, therefore, consisted of twenty animals each, well matched as to age and growth tendency, and perhaps to a lesser degree, as to certain temperamental or personality characteristics.

Approximately eight months later, measurements of liquid intake under different conditions of availability of food were begun. The average daily liquid intake of the salt and water groups was determined over a 2-week period, with food freely available. Both groups consumed approximately 12–14 gm. of food daily during this time. Thereafter the animals were placed on a deprivation schedule designed to reduce their weight and then maintain it at the lowered level. To accomplish this, 5 gm. of food were given each animal daily for 4 days. During the next three days they received only enough food (1–8 gm.) to reduce their weight to 85 per cent of the weight at the beginning of the determinations[13]. During the next three weeks the weight of all rats was kept constant, within a daily average fluctuation of ± 2 gm. per group, by supplying them with an average ration of 9 gm. per day. Determinations of liquid intake during the periods of weight reduction and maintenance were made. Thereafter the animals were given food *ad libitum* and later used for other studies.

As seen from Table 1, the average daily fluid intake of the group drinking only water remained almost constant during the three different periods of food availability. The salt group behaved differently. As expected, these animals, when offered a choice between tap water and a 1 per cent salt solution, strongly preferred the latter under all conditions. Their fluid intake under ordinary conditions, that is, with food freely available, was significantly higher than that of the water group ($t = 4·82$; $P < 0·001$). During both periods of food restriction the average daily intake of fresh water in the salt group did not vary significantly from the volume consumed during the period of free access to food. The consumption of 1 per cent salt solution, however, rose markedly, so that the average number of c.c. drunk daily during the seven days of weight reduction and the fourteen days of maintenance of reduced weight was almost twice that consumed under ordinary cage conditions.

Thus the results are clear in indicating that when rats are deprived of their normal ration of food their consumption of water does not change, but when given the opportunity, their consumption of a 1 per cent salt solution will rise steeply, so that their average daily total fluid intake is almost three times (Table 1) that of animals drinking water only.

The average daily food ration necessary to maintain the animals at the desired level of weight reduction (85 per cent of initial weight) varied from 8·6 ± 0·3 to 10·4 ± 0·4 gm., with a mean ration, as mentioned earlier, of 9 gm. in both the experimental and control groups. This suggests that the increased intake and excretion of salt and water were carried out efficiently by the salt solution drinkers, since additional food or caloric intake was not required by this group.

After one year of observation, no significant difference between salt and water drinkers was noted in survival, growth (gain in weight), or fertility (Table 2). A separate group of twenty female rats, maintained on a free choice of 1 per cent salt solution and/or water for one year, but not included in any of the other studies, proved also to be fertile. Litters from male and female salt drinkers, each mated with a fresh-water drinker of the opposite sex, were fully the equivalent of control progeny in number, survival, and weight at weaning. It is not impossible, however, that differences in survival and fertility between salt and water drinkers would have become apparent had the observation period been carried beyond one year.

Determinations of the weights of heart ventricles and kidneys after food deprivation were made on two additional series of rats, not included in the fluid intake studies. To provide a background of organ-weights in our rat colony against which to evaluate these findings, the organ-weights of 427 normal animals, ranging in weight from 60 to 700 gm. were determined. A log–log plot of organ versus body-weight gives a positive straight-line relationship between the two over wide ranges of body-weight, with a uniform spread (standard deviation) about

Table 2 WEIGHT, SURVIVAL AND FERTILITY OF MALE RATS OFFERED A 1 PER CENT SALT SOLUTION AND WATER, OR WATER ALONE, AFTER ONE YEAR OF OBSERVATION

Group	No. of rats	Average weight (gm.) ±standard error	No. alive		No fertile
Salt solution and water	36	491±22	26	$\chi^2 = 2·432$ df = 1 $P > 0·10$	26
Water only	32	486±17	28		28

These groups are not identical with the animals used in the fluid intake studies, since a number of those were killed for other determinations. Comparable male animals, selected and maintained in the same way as were the fluid intake groups, but not subjected to the deprivation schedules, have been included instead.

Table 3. Analysis of the Effect of Food Restriction upon Heart and Kidney Weights in Rats Drinking 1 per cent Salt Solution and Water, or Water only

Group	No. of rats	Average weight at death (gm.)	Average weight of heart ventricles (gm.)		Average weight of both kidneys (gm.)	
Salt solution and water	30	293 ± 4·8*	0·73 ± 0·014	$t = 4·49$ $P < 0·001$	1·9 ± 0·05	$t = 3·12$ $P < 0·002$
Water only	30	291 ± 4·7	0·65 ± 0·011		1·7 ± 0·04	

The six groups of ten rats each, seen in Fig. 1, have been re-grouped into two series of thirty each for computational purposes. Animals were killed at end of period of maintenance of reduced weight, at 124 days of age.

* Standard error of the mean.

each mean weight[14]. In Fig. 1, the solid lines represent the normal relationships of heart ventricle and kidney weights to body-weights, over the body-weight range of the animals in the current experimental series. The dotted lines represent the standard deviations of the plotted mean organ weights.

Experimental results are presented in Fig. 1 as points, each of which represents the mean organ weight of ten animals, killed directly after termination of the period of maintenance of reduced weight, at 124 days of age. The salt animals were offered a choice of 1 per cent salt solution or water from weaning (24 days), while the others received water only. It can be seen that the effect of food restriction was to reduce the average weights of ventricles and kidneys of both salt and water drinkers, as compared with colony control results, but the degree of weight loss of both organs was significantly less in the salt solution group than in the straight water drinkers (Table 3). A second series (not shown in Fig. 3) of 50 salt solution drinkers and 53 water-drinking rats was kept on the food deprivation and maintenance of reduced weight schedule already described, and then permitted to eat freely for 3–16 weeks thereafter. No deviations of their ventricle and kidney weights from those of the colony controls were found. Both groups ranged, in age, from 84 to 455 days, and in average weight from 310 to 512 gm., at the time they were killed for determination of organ weights.

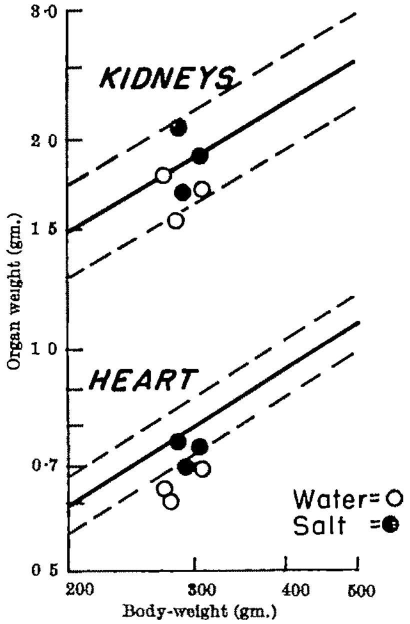

Fig. 1. Effect of food restriction on weight of heart ventricles and kidneys in rats drinking 1 per cent salt solution and water, or water only. Each point represents the average organ-weight of 10 animals. The solid lines are log–log plots of organ- vs. body-weights of normally reared animals; the broken lines represent the standard deviations of these plots

No differences in the weights of the adrenal glands, livers or thymus glands were noted between the salt drinkers and the rats drinking water. Under food restriction the weight of the adrenals increased by about 20 per cent in both groups, and that of the liver and thymus gland decreased by 30 and 50 per cent, respectively. In the groups that were given free access to food for 3–16 weeks following reduced food intake, the weights of these organs closely paralleled normal values.

Taken together these results document certain consequences (or the lack thereof) of freely chosen, long-term ingestion of salt. Consumption of 1 per cent salt solution for periods of more than a year had no effect upon the weight of the heart ventricles or kidneys of normally reared rats. Under conditions of food restriction the loss of weight of these organs was significantly reduced in the presence of salt intake. Survival and body-weight were not affected after one year of drinking 1 per cent salt solution, nor was the fertility or the quality or number of offspring of such animals altered.

During the period of reduced weight maintenance the salt and water groups both required the same amount of food to maintain their weights. Yet the animals given a free choice of 1 per cent salt solution or tap water increased their average daily fluid intake (consisting overwhelmingly of 1 per cent salt solution and *not* water) to 25 per cent of their body-weight (Table 1), while the intake of the water group remained stable.

We believe these results provide an example of rapid learning in the rat, where a lack (food deprivation) in one aspect of consummatory behaviour (eating) leads to a marked change (increased intake of salt solution) in another consummatory response (drinking), in the presence of adequate neural equipment, and experience (rapidly acquired preference for 1 per cent salt solution). The rat's preference for 1 per cent salt solution is manifested quickly, and is maintained consistently under normal cage conditions. (We do not believe it was necessary to give the animals a free choice of salt solution or water from the time of weaning in order to demonstrate these findings. The long-term intake of salt solution was permitted only for the purposes of the toxicity studies.) When the amount of food available to the rat was curtailed, its intake of another appetitively stimulating substance, salt, increased, but only to a certain level. These animals were not starving, and presumably the maximal satiating effect of salt solution was reached at that level.

This interpretation is supported by the results in Table 1, which indicate that during the 7-day period of weight reduction, the average daily intake of salt solution was 59 c.c.; but during the next 21 days of maintenance of reduced weight, when the animals actually were receiving a greater amount of food than before, though still less than under *ad libitum* feeding, the intake of salt solution became stabilized at 65 c.c. Thus the consumption of salt solution was approximately the same under both conditions of food reduction, but with ample food available, it was markedly lower (34 c.c. a day). That this effect is transitory and related to the availability of food was further shown by observation of the rats after their return to *ad libitum* feeding, when their average intake of salt solution dropped to initial levels.

Hebb has presented a theoretical account of the role of learning in hunger, eating, and the cessation of eating[15]. He illustrates some of his points with the salted nuts phenomenon, where once they are tasted, it becomes increasingly difficult to let them alone. Hunger has increased, but how? As Hebb says, ". . . a lack of food cannot be increased by eating something, and stomach contractions are stopped by chewing and swallowing" (ref. 15, p. 200). Nor is it feasible to believe that a taste of salted nuts increases appetite and not hunger, for as Hebb points out, the distinction between the two, at least in terms of an understanding of the physiological control of eating, is highly artificial. If, however, one considers hunger, ". . . to be neither a particular condition of the body, nor a set of sensations from the stomach, but an organized neural activity that can be aroused (like any other conceptual process) in several ways . . ." (ref. 15, p. 203), the role of salt as a provocative agent in consummatory activity is somewhat clarified. Drinking only tap water evidently did not provide an associative link for the animals, in these experiments, between the consummatory response made and appetite. Put another way, it did not serve (as did salt) to arouse a pattern of learned behaviour associated with appetite and/or hunger.

The recognition of the role of learning or experience in activities generally viewed as responses to internal needs, or primary biological drives, will require a re-appraisal of current ideas about hunger and thirst, and perhaps of greater significance for behaviour theory, of the concept of motivation, in general. The increasing recognition that the functional division of the nervous system into 'central' and 'peripheral' (or 'sensory') mechanisms is probably unwarranted with regard to behaviour should be of particular pertinence in this connexion[16]. Recent work by a number of investigators[17-19], as well as a current review[20] and a book[21], emphasizing the role of experience in the determination of patterns of consummatory behaviour, lend additional support to these conclusions.

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[1] Dahl, L. K., *Nature*, **181**, 989 (1958).
[2] Bass, D. E., and Kleeman, C. R., Quartermaster Climatic Res. Lab. Rep. No. 214, Lawrence, Mass. (1958).
[3] Loeb, R. F., *Proc. Soc. Exp. Biol. Med.*, **30**, 808 (1933).
[4] Blum, L., and Grabar, P., *C. R. Soc. Biol.*, **98**, 527 (1928).
[5] Kaunitz, H., *Nature*, **178**, 1141 (1956).
[6] Richter, C. P., "L'Instinct dans le Comportement des Animaux et de l'Homme" (Masson, Paris, 1956).
[7] Bare, J. K., *Amer. Psycholog.*, **2**, 331 (Abst.) (1947).
[8] Weiner, I. H., and Stellar, E , *J. Comp. Physiol. Psychol.*, **44**, 394 (1951).
[9] Young, P. T., and Falls, J. L., *J. Comp. Physiol. Psychol.*, **49**, 569 (1956).
[10] Kaunitz, H., *Wien. Med. Wochschrift.*, **102**, 356 (1952).
[11] Lashley, K. S., "Brain Mechanisms and Intelligence" (Univ. Chicago Press, Chicago, 1929).
[12] Hall, C. S., *Psychol. Bull.*, **38**, 909 (1941).
[13] Kaunitz, H , Slanetz, C. A., Johnson, R. E , and Guilmain, J., *J. Nutrition*, **60**, 221 (1956).
[14] Stoerk, H. C., and Zucker, T. F., *Proc. Soc. Exp. Biol. Med.*, **6**, 297 (1946).
[15] Hebb, D. O., "The Organization of Behavior. A Neuropsychological Theory" (Wiley and Sons, New York, 1949).
[16] Teuber, H. L., "Ann. Rev. Psychol.", **6**, 267 (1955).
[17] Ghent, L., *J. Comp. Physiol. Psychol.*, **50**, 172 (1957).
[18] Lawrence, D. H., and Mason, W. A., *J. Comp. Physiol. Psychol.*, **48**, 267 (1955).
[19] Baker, R. A., *J. Comp. Physiol. Psychol.*, **48**, 37 (1955).
[20] Cofer, C. H , "Ann. Rev. Psychol.", **10**, 173 (1959).
[21] Bindra, D., "Motivation: A Systematic Reinterpretation" (Ronald Press, New York, 1959).

NEWS and VIEWS

Radio Research in the D.S.I.R.: Dr. R. L. Smith-Rose, C.B.E.

On September 30, Dr. R. L. Smith-Rose will retire as director of radio research in the Department of Scientific and Industrial Research. Dr. Smith-Rose has been associated with the work of the Radio Research Board of the Department since the formation of the Board in 1920. In 1919 he joined the scientific staff of the National Physical Laboratory as a member of the Electricity Department, and a few years later, when the new 'Wireless Division' of the Department was formed, Smith-Rose was appointed scientific officer-in-charge. This was the nucleus of the Radio Division of which he became superintendent in 1939. In 1948 he was appointed to the new post of director of radio research in the Department of Scientific and Industrial Research, with headquarters at the Radio Research Station, Slough. This Research Station is now known internationally for its work in the fields of radio wave propagation, tropospheric and ionospheric research, radio atmospheric noise, radio standards and measurements and, more recently, for observations and predictions on artificial Earth satellites. Smith-Rose's personal research activities have included fundamental and pioneering investigations in radio direction finding, studies on the electrical properties of soil and sea water, and on the propagation of radio waves over the ground and in the troposphere. He has always been very active in national and international radio organizations and has held numerous posts of distinction. He was chairman of the Radio Section of the Institution of Electrical Engineers in 1943, and has been awarded premiums on five occasions for original papers read before the Institution. He is a Fellow and former vice-president of the Institute of Radio Engineers (New York) and is an honorary member of the Institution of Radio Engineers, Australia. He is a vice-president of the International Scientific Radio Union. In 1947, Dr. Smith-Rose was awarded the U.S. Medal of Freedom with Silver Palm for his service in scientific research and development, and for co-operation with American scientists. He was made C.B.E. in 1952.

Mr. J. A. Ratcliffe, C.B.E., F.R.S.

MR. J. A. RATCLIFFE has been appointed to succeed Dr. Smith-Rose. At present Mr. Ratcliffe is reader in physics in the University of Cambridge and head of the Radio Section at the Cavendish Laboratory. He entered Cambridge in 1921 with an open scholarship to Sidney Sussex College and first joined the Cavendish research group in 1924, working with Appleton in some of his earliest investigations of the ionosphere. In 1927 he was appointed demonstrator at the Cavendish Laboratory and in the same year elected a Fellow of Sidney Sussex. During the War, Ratcliffe was employed on various aspects of research and development in radio, mainly at the Telecommunications Research Establishment, Malvern. For a year he was seconded to the Army to start the radar school at Petersham and later returned to the establishment to undertake responsibility for establishing and running the Post-Design Service organization which was concerned with performance problems of radar equipment under service conditions. In 1947 he was made O.B.E., for this war-time work, and in 1959 C.B.E., for advisory work to the Ministry of Supply. In 1951 he was elected a Fellow of the Royal Society, and at the present time he is president of the Physical Society, vice-chairman of the Electronics and Communications Section of the Institution of Electrical Engineering, and overseas vice-president of the Institute of Radio Engineers (New York). Apart from the war years, Ratcliffe's work has been carried out at Cambridge where he has built up a major radio research school. Over a period of thirty-five years he has made extensive researches on the propagation of radio waves and on the ionosphere.

U.K. Atomic Energy Authority : Mr. H. Cartwright, M.B.E.

MR. H. CARTWRIGHT, at present serving as deputy director of industrial power in the Development and Engineering Group of the United Kingdom Atomic Energy Authority at Risley, has been appointed director of industrial power. Mr. Cartwright was born in 1919 in Manchester. He was educated at William Hulme Grammar School, Manchester, and at St. John's College, Cambridge, where he took a first-class honours degree in the Mechanical Sciences Tripos in 1940. During the period 1942–46 he was a signals officer on ground radar stations in the Royal Air Force, reaching the rank of flight lieutenant. After working for the Decca Navigator Co., Ltd., Brixton, and the English Electric Co., Ltd., Stafford, he joined the Atomic Energy Division of the Ministry of Supply at Risley in 1949 as a design engineer. In 1954, on the formation of the Atomic Energy Authority, Mr. Cartwright became an assistant chief engineer in the Authority's Industrial Group at Risley. He was promoted to chief engineer in 1955, assistant director in 1956 and deputy director of industrial power in 1958.

International Atomic Energy Agency: Research Grant

THE first research grant under the Exchange and Training Programme of the International Atomic Energy Agency has been awarded to Dr R. P Agarwala. Dr. Agarwala has graduated from the Agra College, India, and the Imperial College of Science and Technology in London; he has published a number of papers on metallurgy Dr. Agarwala will undertake a year of research in the field of solid-state physics and chemistry at the Massachusetts Institute of Technology. These research grants are awarded mainly to scientists with considerable research experience, who are already working in their own countries on a promising line of research which cannot be developed fully there because of lack of equipment or laboratory facilities.

Silver Jubilee of *Science and Culture*

THE Silver Jubilee issue of *Science and Culture* (25, 1), organ of the Indian Science News Association, is one of which its sponsors may justly be proud. Besides a brief and modest history of the Association itself, there is an account of the rise of scientific research in India with tributes to those pioneers who did so much in difficult times. Among the contributors to the Silver Jubilee edition are distinguished men of science from various parts of the world. Hans von Euler, of the University of Stockholm, examines experimental work on nuclear acids and nucleotides, and A. I. Oparin, of the Moscow Academy of Sciences, discusses the origin and development of life on Earth. J. D. Bernal considers priorities and phasing in the use of science, while Ivan Matels, of the Czechoslovak Academy of Science, describes research and production of antibiotics in Czechoslovakia. There are also a number of articles by eminent men of science from India. The journal is published monthly from 92 Achanya Prafullachandra Road, Calcutta 9, at an annual subscription rate of £1.

Alcohol and Road Safety

ALTHOUGH the problem of alcohol and road safety has been much in the public eye, there has hitherto been little precise evidence as to the effects of small quantities of alcohol on driving skill. For this reason, the careful study by Prof. George Drew and his colleagues recently published by the Medical Research Council ("Effect of Small Doses of Alcohol on a Skill Resembling Driving", by G. C. Drew, W. P. Colquhoun and Hazel A. Long M.R.C. Memorandum No. 38. H.M.S.O. 7*s*. 6*d*. net) will attract widespread interest. This report describes the effects of small doses of alcohol (the highest being roughly equivalent to only five fluid ounces of whisky for a man of average weight) upon performance on the 'Miles motor driving trainer'—a complicated tracking task which has a good deal in common with actual driving. It was found that the mean error of forty subjects on the test increased significantly with increasing blood alcohol, the shape of the curve being strikingly similar to the blood and urine alcohol curves On the other hand, the average speed of 'driving' showed no significant change. Marked differences between subjects were however observed, and the authors rightly place weight on temperamental factors as governing individual reactions to alcohol.

These results strongly suggest that amounts of alcohol far too small to give rise to recognizable signs of intoxication may none the less significantly impair driving ability. At the same time, the authors are properly cautious in generalizing from their findings to driving—and drinking—under conditions less austere than those of the laboratory. The effects of small doses of alcohol upon confidence, decision and judgment were assessed little, if at all, in the present series of experiments, and the range and extent of compensatory activities have still to be defined with precision. None the less, this report deserves the most careful attention on the part of all concerned with the pressing problem of road safety.

Domestic Oil-heating

"BRITAIN Turns to Oil Heat" is the title of an anonymous but significant contribution to the combined summer and autumn (1959) number of the *Esso Magazine*, house organ of the Esso Petroleum Co. Domestic heating by oil is increasing rapidly. Just after the War there were 5,000 houses with oil-fired central heating; in 1957 there were 16,000; and in 1958, 40,000. Similarly, portable oil heaters have increased in popularity from 2½ million to more than 10 million in the past five years Perhaps the most interesting information in this article, especially in view of recent publicity given to fatal accidents with oil heaters, is quoted from a consumer's association. Ten different models were burned together in a room for several hours without creating odour. They were then knocked on their backs to see if they would spill burning oil; but in every case the flame either went out or returned to normal burning when the heater was set up again in correct position. Finally, the models were subjected to blasts of air to see whether flames would burn in a dangerous manner. In no case did anything catch fire, a remarkable result and tribute to the various heater designs involved.

Parametric Amplifier Diode

'EW97/1', a variable capacitance diode designed for use in parametric amplifiers at frequencies up to *S*-band, is now in production by the General Electric Co., Ltd. (Semiconductor Division). The Company claims that it is the first device with these qualities to be made available in the United Kingdom. The price is considerably lower than that of the units which previously had to be imported. The diode is suitable for use in radar and other communications systems. Designed for high power dissipation, it needs no elaborate precautions against surges. It is mounted in a coaxial structure for direct insertion into coaxial and wave-guide circuits and it has the very low series inductance of 0·5 μH. A reverse polarity version ('EW98/1') is also available. Because of its very low forward impedance and very high reverse impedance, the device can be used as a micro-wave switch. Another application is as a frequency multiplier—an important aid towards the future design of micro-wave receivers based entirely on solid-state devices.

Palæolithic Remains at Willendorf

EVERY prehistorian knows of the Venus of Willendorf, and some of them know that the figurine comes from a site near the village of the same name which lies on the left bank of the Danube between Melk and Krems, upstream from Vienna. The figurine itself is made of a fine-grained oolite, is some 10 cm. in height, and is highly conventionalized. It can be dated to a late palæolithic epoch. But up to now very little detail has been generally known about the find, the site itself, and the associated industry. Now three fascicules of the Prehistoric Commission of the Austrian Academy of Science have been devoted to these matters (Mitteilungen der Prähistorischen Kommission der Österreichischen Akademie der Wissenschaften. VIII und IX Band: Willendorf in der Wachau: Monographie der Paläolith-Fundstellen I–VII. 1 Teil: Text. Pp. v+219. 2 Teil: Inventar. Pp. 79. 3 Teil: Abbildungen. Abb. 124. (Wien: Österreichische Akademie der Wissenschaften, 1959)). At Willendorf there are a number of loess sites which have yielded upper palæolithic industries; the famous figurine came from site 2. The first fascicule includes a full account of the sites and the various excavations that have taken place there; Part 2 is a catalogue of all the finds of stone and bone tools that have been unearthed; Part 3 is entirely taken up with pictures of the more important tools, which enable datings for the various levels to be suggested. A work of this nature is long overdue. Now at last prehistorians have the full facts connected with one of the more important works of art of late palæolithic man that has survived.

The Transvaal Museum

THE annual report of the Transvaal Museum, Pretoria, for the year ended March 31, 1959, once again directs attention to the desperate need for more accommodation The Trustees consider that the people of South Africa have been unduly penalized in that large sections of the collections in both the natural sciences and history are steadily deteriorating owing to lack of storage and display space. Proposals for a separate and independent cultural history museum and an open-air historical museum are also being discussed at a high level with the Minister of Education. An entrance charge (1*s*. per head for adults but free for children under sixteen and recognized groups of students) has been made. The revenue is devoted exclusively to the improvement of the collections, and it is interesting to note that the decrease in attendances is not nearly so great as was expected.

Student Exchange

DURING the summer vacation of 1959 the number of students sent and received under the programme of the International Association for the Exchange of Students for Technical Experience of Great Britain increased to 854 students received, and 837 students sent; the corresponding figures for 1958 were 829 and 774, respectively. Some 366 British companies took part in the scheme. In April 1959 a general meeting of representatives of affiliated universities, colleges and companies was convened to consider the future organization of the Association, in Great Britain. It was decided to establish an independent council to control the Association, and the legal formalities are now proceeding. It is anticipated that the new Council of the Association will operate from September 1960. Greece has been accepted as a full member of the Association, and Poland and Tunisia have been admitted as associate member countries for one year. Further information about the Association can be obtained from J. Newby, superintendent of vacation studies, Imperial College of Science and Technology, London, S.W.7.

Scottish Field Studies Association

THE Scottish Field Studies Association has announced its programme for the forthcoming season April–September. The courses will be based on the Garth Field Centre and the Isle of Ramsey, and accommodation fees will be up to 4½ guineas a week. Members of local scientific societies qualify for bursaries of £3 (for a course lasting a week) from a fund made available by the Carnegie United Kingdom Trust. Further information can be obtained from the Scottish Field Studies Association, c/o Department of Botany, The University, Glasgow, W.2.

Symposium on Herbicides and the Soil

The British Weed Control Council has arranged a symposium on Herbicides and the Soil, which will be held in the Clarendon Laboratory, Parks Road, Oxford, on April 7. Four papers will be presented. The first will be by Prof. L. J. Audus (Bedford College, London) on "Microbiological Breakdown of Herbicides in Soils". Prof. W. Fletcher (West of Scotland Agricultural College, Glasgow) will speak on "The Effects of Herbicides on Soil Micro-Organisms", and Dr. G. S. Hartley (Chesterford Park Research Station) on "Physico-chemical Aspects of the Availability of Herbicides in Soils". The symposium will be concluded by Dr. Ir. W. van der Zweep (Wageningen), who will speak on "Persistence in the Soil of Some Important Herbicides". Further information can be obtained from G. R. Sagar, Unit of Experimental Agronomy, Department of Agriculture, University of Oxford.

Announcements

Dr. D. A. Senior, whose appointment to the newly created post of scientific attaché to the British Embassy in Moscow was announced in *Nature* of April 11, 1959, p. 1018, left London to take up his post on January 31. Dr. Senior has no specific technical assignment in Moscow, but will cover the broad field of science and technology. His aim is to promote good relations and understanding between the two countries in this field and to facilitate exchanges of information and people.

Dr. A. D. Macdonald, Leech professor of pharmacology in the University of Manchester, has been appointed Officier de l'Ordre de la Santé Publique by the French Minister of Health. Prof. Macdonald, one of the members of the Pharmaceutical Society's Council nominated by the Privy Council, has been for two years president of the British section of the Franco-British Pharmaceutical Commission, a body set up as a result of a suggestion made by Sir Winston Churchill that members of the same professions on each side of the Channel should get to know one another better.

Dr. A. W. Meschkat, of Hamburg, has been appointed as Food and Agriculture Organization regional fisheries officer for Africa. He will assist governments to develop fish production of the seas off East and West Africa and of the lakes and rivers of Central Africa.

Mr. A. L. Jewell, assistant curator of Haslemere Museum, has been appointed to a newly created post of exhibitions officer at the Royal Botanic Gardens at Kew. Mr. Jewell will be joining the staff of the Keeper of Museums at Kew and his special concern will be to re-display existing exhibits.

Dr. R. H. Hedley, a senior scientific officer in the Department of Zoology, British Museum (Natural History), has been awarded the 1960 New Zealand National Research Fellowship and will work on some aspects of the biology and systematics of marine allogromoid Foraminifera (Protozoa) living in New Zealand coastal waters.

The Corday-Morgan Award, consisting of a silver medal and a monetary prize of 200 guineas, is made annually to a chemist of British nationality who, in the judgment of the Council of the Chemical Society, has published during the year in question the most meritorious contribution to experimental chemistry, and who has not, at the date of publication, attained the age of thirty-six years. Copies of the rules governing the Award may be obtained from the General Secretary of the Society. Applications or recommendations in respect of the Award for the year 1959 must be received not later than December 31, 1960, and applications for the Award for 1960 are due before the end of 1961.

The sixty-fifth annual congress of the South-Eastern Union of Scientific Societies will be held at Ipswich during April 22–25. Further information can be obtained from Mr. Frank Edwards, 53 The Drive, Shoreham-by-Sea, Sussex.

The annual spring conference of the Institute of Metal Finishing will be held in the Grand Hotel, Scarborough, during April 26–30. Further information can be obtained from the Honorary Secretary, Institute of Metal Finishing, 32 Great Ormond Street, London, W.C.1.

The spring conference of the X-Ray Analysis Group of the Institute of Physics will be held in the main Chemistry Lecture Theatre, University of Reading, during April 8–9. The subject will be "The Borders of X-ray Analysis". Further information can be obtained from Dr. P. T. Davies, "Shell" Research, Ltd., Thornton Research Centre, P.O. Box 1, Chester.

A symposium, sponsored by the Office of Ordnance Research U.S. Army, will be held in the Inorganic Chemistry Laboratory, Oxford, during April 5–6, on Plastic Crystals and Rotation in the Solid State. Further details can be obtained from L. A. K. Staveley, Inorganic Chemistry Laboratory, South Parks Road, Oxford.

The Ergonomics Research Society is holding its annual conference at the University of Cambridge during March 28–31. The programme will include two sessions on "Ergonomics in the Armed Forces", a session on "Ergonomics in the Netherlands", and a symposium on "Factors in Prolonged Work, especially Inspection". Further information can be obtained from the conference secretary, Dr. S. Griew, University of Bristol, Department of Psychology, 22 Berkeley Square, Bristol 8.

The Turkish Bibliography of Articles appearing in Turkish Periodicals for January–March 1959 is provided with a foreword in French as well as Turkish, and the classification, too, is now given in French as well as in Turkish. Titles of the periodicals covered are listed in French and Turkish, and each entry is provided with a French translation of the title; titles are arranged according to the Dewey Decimal Classification up to the first two or three figures only.

A research scholarship in the use of light alloys in structural engineering is being offered by the Institution of Structural Engineers in collaboration with the Aluminium Development Association. The duration of the scholarship will be for two years and the value £600 per annum. The award is made every three years as from October 1. Further information can be obtained from the Secretary of the Institution of Structural Engineers. Completed application forms should be sent to reach the Secretary by March 31.

INDUSTRIAL ARCHÆOLOGY

THE conference held at the London School of Hygiene and Tropical Medicine on December 12 was the first event organized by the recently formed Research Committee on Industrial Archæology of the Council for British Archæology. The large attendance of some 180 people was very encouraging, and confirms the Council in its belief that considerable interest exists as well as concern about the fate of industrial monuments.

Dr. J. N. L. Myres, president of the Council, took the chair, and the conference opened with the reading of a message from the Minister of Works. The first paper was by Mr. M. M. Rix, staff tutor in architectural history in the University of Birmingham, on "Industrial Archæology as illustrated in the West Midlands". This area is a particularly rich field, and Mr. Rix described many fine examples such as Ironbridge and Coalbrookdale, Telford's canal aqueducts, and the earliest existing iron-framed building, a former flax-spinning mill at Shrewsbury.

The second paper, by Mr. J. M. Palmer, was on "The Stamford Canal: a Seventeenth Century Navigation". His chief purpose was to show how the history of a canal, the very site of which can only with difficulty be identified, may be reconstructed by a thorough search of national and local records. The paper was introduced by Mr. Maurice Berrill, honorary secretary of the Railway and Canal Historical Society.

The afternoon session began with a paper by Prof. A. W. Skempton, of the Imperial College of Science and Technology, London, on "Structural Development of the First Iron-framed Textile Mills". He showed how iron-framing was originally developed because of the danger of fire in early factory buildings. The development was one of very great importance as it culminated in the steel-frame buildings of the first examples of modern architecture designed by Louis H. Sullivan in Chicago towards the end of last century. Prof. Skempton showed how skilfully he had combined study of actual buildings and documentary records to establish the sequence of mills in Derbyshire and at Shrewsbury and Salford.

The remainder of the afternoon session was devoted to three short papers. The first was a brief note by Mr. C. W. Phillips, read in his absence owing to illness, on the Ordnance Survey in relation to the recording of industrial archæology. Mr. Phillips, who is archæology officer to the Ordnance Survey, pointed out that surveying and recording are being extended to include early industrial remains. He also made it clear that the records compiled by the Ordnance Survey on such sites will be available to research workers. Dr. P. M. G. Eden, of the Royal Commission on Ancient Monuments, gave a most practical short address on the technique of recording industrial monuments. He encouraged amateur field groups by emphasizing that preliminary work in recording often serves as a basis for specialist studies. Mr. J. M. Richards, editor of the *Architectural Review*, read the final paper on "Visual Aspects of the Subject and the Æsthetic and Landscape Value of Early Industrial Monuments", in which he distinguished between the architectural quality of industrial buildings and their part in the landscape in so far as it was a creation of man. Mr. Richards's thesis that the best of our early industrial buildings mark the beginning of the functional tradition in architecture is already well known.

In the discussions which were held after the papers during both morning and afternoon sessions a keen concern could be observed with how the recording of industrial remains might be carried out and appropriate examples preserved. The need for the co-ordination of the activities of the different societies working in the field of industrial archæology was emphasized, as was the necessity for an informed public opinion which could bring pressure to bear on the appropriate authorities. It was even suggested that the Royal Commission on Historical Monuments should appoint additional staff to deal with the survey of industrial remains. Hopeful developments were referred to such as the action of Allied Ironfounders in clearing the site of the old Coalbrookdale ironworks, the first scientific excavation of an industrial site by the British Museum at the Longton Hall porcelain factory, and the appointment of an archæological officer in the Staffordshire County Planning Department.

The conference closed with a summary by Mr. Maurice Barley, secretary of the Council for British Archæology, at the conclusion of which he read a draft resolution. A discussion followed in which the chief points made were the necessity to interest industry in the work of record and preservation and to emphasize the educational importance of industrial archæology when so much was being heard of the need for increased technical and scientific training.

The resolution as finally passed declared that the conference was deeply impressed by the need for formulating a policy for recording and, where necessary, preserving early industrial monuments. The Research Committee on Industrial Archæology was further instructed by the resolution to find opportunities to discuss with central and local authorities, as well as with industry and unofficial bodies, the question of record and preservation.

The Research Committee proposed as a first step to prepare a handbook of industrial archæology which would serve as a guide to societies and individual research workers anxious to carry out surveys of industrial remains. Plans are now being made to initiate a pilot survey either of a region or of a particular industry. It is also hoped to hold regional conferences, and as soon as possible to organize courses at which some training in recording and interpreting industrial sites can be given.

The immediate task of the Committee is to establish closer contact between the individual research workers and societies which have been working in different parts of the field covered by industrial archæology and to attempt to direct their efforts towards the creation of a single discipline. The work of the Committee, if successful, will go a long way towards establishing the study of the history of technology on a more respectable footing in Britain as well as supplementing the work of economic and social historians. Much depends on the degree of interest which can be aroused among scientists and technologists, without whose assistance it will be virtually impossible to go on to specialized surveys of particular industries. The Committee already includes archæologists, historians, architects, geologists and technologists, but would warmly welcome the co-operation of more people with specialized knowledge of mining, quarrying, textiles, metallurgy, chemicals, civil and mechanical engineering or any of the other innumerable subjects in this vast field of industrial archæology. E. R. R. GREEN

NINTH CANADIAN HIGH POLYMER FORUM

THE ninth Canadian High Polymer Forum, held at the Guild Inn, Toronto, during October 26–28, was attended by 142 chemists from Great Britain, the United States and Canada. The chairman was M. H. Jones (Ontario Research Foundation) and the session chairmen were S. Bywater (National Research Council), H. H. G. Jellinek (Essex College), M. Rinfret (University of Montreal) and G. Olah (Dow of Canada).

At the business meeting on October 26 the officers for the tenth Forum were elected: *chairman*, L. A. McLeod, Polymer Corporation; *programme chairman*, K. E. Russell, Queen's University, Kingston, Ontario; *secretary-treasurer*, D. A. I. Goring, Pulp and Paper Research Institute.

At the banquet on October 27, the guest speaker, C. H. Bamford, of Courtaulds, Maidenhead, spoke on "Patterns in Reactions and Polymers". He said that detailed information is now available concerning transfer reactions of polymer radicals with a variety of substrates. For a hydrocarbon substrate such as ethylbenzene, the rate constants for the hydrogen abstraction process involving a series of polymer radicals are fairly accurately proportional to the rate constants for the corresponding reactions with toluene. In these reactions with hydrocarbon substrates, polar factors are very small. With ferric chloride, butyl mercaptan or carbon tetrabromide as substrates, a logarithmic plot of rate constants for the transfer reaction against rate constants for the corresponding reaction with toluene gives a number of scattered points. The pattern of points is, however, very much the same in all three cases. With triethylamine as substrate an 'inverse' pattern is obtained. Patterns of the first type are obtained in addition reactions involving the same polymer radicals and a number of vinyl monomers.

The observed patterns can be interpreted in terms of polar effects, the rate constant depending considerably on the extent to which the substituent in the radical is electron-withdrawing or electron-releasing. From a quantitative treatment, Dr. Bamford concluded that it is possible to express the rate constant for the transfer reaction of a radical with a given substrate in terms of the rate constant for transfer with toluene and the Hammett σ constant for the radical. Predictions based on this quantitative theory are in excellent agreement with experiment.

The main sessions began with a paper by J. K. N. Jones (Queen's University) on "Some Problems in Polysaccharide Chemistry". In pointing out some of the difficulties involved in complete structural determinations of polysaccharides, he said that it is not always possible to decide whether traces of sugars occurring with the main hydrolysis products of polysaccharides were part of the original macromolecule. Degrees of branching can be readily obtained using the Haworth methylation technique, but frequently it is almost impossible to achieve 100 per cent methylation of a polysaccharide. The major problem in this field is to imitate the action of enzymes on sugars and sugar derivatives to produce polymers similar to those found in plants and animals.

B. L. Funt and F. D. Williams (University of Manitoba) in their paper on "Retardation and Inhibition of Methyl Methacrylate Polymerization" described investigations of the effect of benzoquinone on methyl methacrylate polymerization using quinone and initiator labelled with carbon-14. They observed a transition from retardation to inhibition at a critical concentration of benzoquinone. The Monday morning session closed with a short colour film on polymer interactions made available by W. F. Busse, Du Pont Co., Wilmington.

The first in a series of papers on polymer degradation was given by H. A. Kraessig and J. Neal (Industrial Cellulose Research), who discussed "The Degradation of Cellulose by Megavolt Electrons". They showed that at low dose-rates the number of chemical bonds broken is a linear function of the dose, and the sensitive volume corresponding to cleavage of a single bond is close to the volume of a monomer unit. Experiments on wet cellulose indicate that water has a considerable protective action. H. H. G. Jellinek (Essex College) and W. A. Schlueter described work performed at the University of Cincinatti on "The Photodegradation of Polyacrylonitrile in Solution", using light of wave-length 2537 Å. The rate of degradation *in vacuo* is proportional to the light intensity and the number of broken bonds is less than 10^{-3} per quantum absorbed. W. C. Schneider (American Cyanamid) spoke on "The Thermal Stability of Polyacrylonitrile". At 240° in the absence of oxygen he observed that the polymer breaks down very rapidly giving a complex mixture of products, but in the presence of oxygen a more controlled reaction gives water, ammonia and hydrogen cyanide as the only volatile products. J. R. Schaefgen (Du Pont) presented a paper on "The Thermal Degradation of Poly-*p*-xylylene". The rate of degradation was measured in a chlorinated aromatic solvent in the range 285–321°. Degradation is random, and the activation energy is 58 kcal./mole. Anthracene decreased the rate of degradation, but common transfer agents are inactive.

R. J. Orr (Polymer Corporation) presented a paper on "Thermochemical Aspects of Free Radical Butadiene-Styrene Copolymerization". A treatment of the thermodynamics of copolymerization has been devised which can be applied to high-conversion material. The sum of the entropy changes for the two heteropolymerization steps was found to be about 50 entropy units/mole assuming that the residual entropy contributed by the glassy state is zero.

C. Booth and L. R. Beason (Shell Chemical) gave a paper on "Statistical Treatment of Polymer Fractionation Data". A styrene-butadiene copolymer was fractionated and the fractionation data subjected to a simple binomial analysis. The resulting distribution of molecular weight was in good agreement with that calculated from the kinetics of the emulsion system. J. A. Manson, W. W. Graessley and L. M. Hobbs (Air Reduction Co.) described "The Determination of the Breadth of Molecular Weight Distribution in Polyethylene by an Irradiation Technique". Samples of high-pressure and low-pressure polyethylene were irradiated using a cobalt-60 source and were analysed for soluble and gel fractions. From the results it is possible to calculate weight and number average molecular weights of the original polyethylene samples.

A. Rezanowich and D. A. I. Goring (Pulp and Paper Research Institute) have investigated "The Polyelectrolyte Properties of a Lignin Sulphonate Microgel". Their measurements of the polyelectrolyte expansion of fractions of sodium lignin sulphonate indicate a microgel structure for the macromolecule. They developed a theory assuming that the microgel structure has free charges only on the surface and showed that it agreed fairly well with experiment.

D. J. Worsfold and S. Bywater (National Research Council) spoke on "Some Properties of Poly-α-methylstyrene produced by Anionic Polymerization". They have made a thorough molecular weight analysis of poly-α-methylstyrene samples produced using sodium naphthenide as catalyst. One important result is the intrinsic viscosity–molecular weight relationship for polymer samples of very sharp distribution.

M. Senez and H. Daoust (University of Montreal), in their paper on "Heat Parameters for Poly*iso*butylene Solutions", compared heat parameters determined for the poly*iso*butylene–chlorobenzene system by calorimetry and by the viscosity method of Fox and Flory. The results fit the Fox and Flory treatment of viscosity data.

W. Heller and M. Nakagaki (Wayne State University), in "The Exact Theory of the Scanning Method for determining Particle Sizes from Light Scattering", gave accurate calculations of the angular positions of maxima and minima for non-absorbing spheres for the large relative refractive index of 1·20. The results for other relative refractive indexes can now be calculated fairly readily.

T. Gillespie (Dow Chemical) presented papers on "The Limited Flocculation of a Colloidal System by a Water Soluble Polymer" and "The Rheology of a Polystyrene Latex thickened with Methylcellulose". His results on the flocculation of styrene–butadiene latex by methylcellulose indicate that the stabilization of a hydrophobic colloid by a hydrophilic colloid is due to changes in the balance of the flocculation and deflocculation processes. Methylcellulose thickens a polystyrene latex by causing a reversible partial flocculation.

A. Novak and E. Whalley (National Research Council) presented data on "The Infra-red Spectrum of Polyformaldehyde" and showed that these data could be interpreted on the assumption of a helical model for the polymer.

E. H. Immergut, G. Kollmann and A. Malatesta (Dunlop Research Centre) reported results on "The Cationic Copolymerization of Propylene and Isoprene". They prepared soluble copolymers under carefully controlled conditions using aluminium chloride as catalyst at −78° (molecular weight range 4,000–10,000). Reactivity ratios for propylene and isoprene are approximately 0·23 and 0·5. M. H. Jones, U. Martius and M. P. Thorne (Ontario Research Foundation) discussed "The Polymerization of 1-Butene by Metal Alkyl–Titanium Halide Catalysts". They reported the results of chemical and X-ray analyses of the complex catalysts produced by the interaction of aluminium triisobutyl or lithium *n*-butyl with titanium trichloride or tetrachloride. Kinetic results for the polymerization of butene-1 using aluminium triisobutyl–titanium trichloride were given ; the products were of very high molecular weight (intrinsic viscosity between 5 and 6).

C. H. Bamford, in a paper on "Termination by Primary Radicals in Vinyl Polymerization", said that when the initiator concentration is high, a plot of rate of polymerization of styrene versus monomer concentration is curved. This can be quantitatively accounted for by assuming termination of some growing polymer radicals by primary radicals from the initiator. The velocity constant for primary radical termination is about sixty times that for mutual termination. The interaction of unlike radicals was also discussed by C. Sivertz and Y. Ebisuzaki (University of Western Ontario) in a paper on "The Measurement of the Cross-Termination Velocity Constants for Picrylhydrazyl and Alkyl Radicals". They studied the thermal decomposition of azobisisobutyronitrile at such low concentrations that the combination of free cyano-alkyl radicals competed with the reaction with diphenylpicrylhydrazyl. The relationship between the two velocity constants was derived.

L. E. Coleman and J. F. Bork (The Lubrizol Corporation) spoke on "The Reactivity Ratios of N-Vinyl Oxazolidone and N-Vinyl Pyrrolidone with Vinyl Monomers". N-Vinyl oxazolidone copolymerizes well with vinyl chloride and vinyl acetate but not with styrene and methyl methacrylate ; N-vinyl pyrrolidone is more reactive than N-vinyl oxazolidone. W. E. Walles, W. F. Tousignant and T. Houtman (Dow Chemical) have studied "Poly-N-vinyl, 5-Methyl 2-Oxazolidone (PVOM) a New Complexing Polymer". PVOM of molecular weight 150,000 is soluble in water below 40° and insoluble at higher temperatures. This behaviour is explained in terms of a change from a structure which presents largely hydrophilic groups to the solvent to one which is almost entirely hydrophobic.

R. J. Ceresa and F. H. Cotton (National College of Rubber Technology, London), in presenting a paper on "The Mechanico-Chemical Modification of High Polymers", said that cold mastication of elastomers in the presence of 0·1 per cent aluminium isopropoxide eventually leads to gels ; under similar conditions a mixture of polyvinyl acetate and polyethylene may give a fair yield of block copolymer.

R. J. Richardson (Atomic Energy of Canada) spoke on "Radiation Grafting of High Polymers". Nylon tenting fabric was irradiated with gamma-rays in the presence of styrene to give polystyrene grafts largely on the surface of the nylon, with a resultant improvement in its weathering properties. Irradiation of nylon and terylene in the presence of acrylonitrile and vinyl pyrrolidone gave a slight improvement in dyeing response.

J. R. Tichy (Maine Medical Center), in his paper on "The Polymerization of Phosphorus Pentachloride and Urea", presented analytical data on the products of this complex polymerization. In the last paper presented, R. W. Lenz and W. K. Carrington (Dow Chemical) discussed "The Preparation of Phenylene Sulphide Polymers by the Macallum Polymerization". The reaction of *p*-dichlorbenzene with sulphur and sodium carbonate at 300–350° to give a high molecular weight phenylene sulphide polymer probably proceeds partly by a direct attack of sulphur diradicals on the dihalide and partly by attack of sodium sulphide formed by reaction of sulphur with sodium carbonate.

K. E. RUSSELL

DIELECTRIC DEVICES

AN informal residential conference on dielectric devices was held in the Electrical Engineering Department of the University of Birmingham during September 14–17, 1959, inclusive, and was attended by representatives from universities, Government establishments and industry. The aims of the conference were to give those participating a broad overall picture of this sector of solid-state physical electronics, to discuss present trends and new ideas connected with the investigation and exploitation of dielectrics, and to stimulate research on dielectric materials and devices. In an attempt to achieve these aims each session was opened with an invited general paper which reviewed physical principles and assessed practical progress in exploitation; each session then continued with shorter papers describing current research on the properties of materials and the mechanisms of devices.

The first paper of the conference opened the session on masers and parametric amplifiers. This was given by I. M. Ross (Services Electronics Research Laboratories, Harlow), who reviewed the mechanisms of these devices and compared their more important characteristics such as gain bandwidth products and noise figures. J. C. Walling (Mullard Research Laboratories, Salfords) then described the paramagnetic resonance characteristics of chromium-doped potassium cobalticyanide and the advantages of maser operation using 'push–pull' and 'push–push' pumping. Experimental realization of both types of double pumping had been obtained in a cavity maser at a signal frequency of 2,900 Mc./s. with respective root-gain bandwidth products of 12 and 19 Mc./s. in the two cases.

In the following paper, by B. Bölger, N. J. Robinson and J. Ubbink (Philips, Eindhoven), read by J. Ubbink, the characteristics of a 1,420 Mc./s. maser were described. A three-level maser using potassium cobalticyanide at 1·4° K. with a pump frequency of 3,850 Mc./s. and a signal frequency of 1,420 Mc./s. had been constructed. A root-gain bandwidth product of 2·7 Mc./s. with a limiting power of 2×10^{-7} W. had been obtained. The final paper of this session dealt with parametric amplification and was given by L. E. Cross (Electrical Research Association, Leatherhead), who considered the use of a ferroelectric material above its Curie temperature as a possible variable reactance for this purpose. The electro-mechanical effects in ferro-electrics were briefly discussed and saturation functions derived for barium titanate and glycine sulphate. Measurements showed that glycine sulphate could not be used at 500 Mc./s. due to enhanced dielectric losses near the Curie temperature.

The second session was opened by G. F. J. Garlick (University of Hull), who reviewed the physical mechanisms of photoconductivity and luminescence. H. G. Lubszynski (E.M.I., Hayes) then explained how the use of an electron beam for making contact to a semi-conductor offers advantages in the case of near insulators because it allows thin films of large cross-sectional area to be used with less danger of shorting. Organic luminescence and the scintillation counter formed the subjects of the next two papers. In the first of these, J. B. Birks (University of Manchester) discussed the fast and slow components of scintillations in organic materials. The former was attributed to direct singlet-state excitation, was subject to primary quenching, and hence depended on the nature of the particle. The latter was attributed to ion recombination leading to excitation of metastable triplet states, was less subject to primary quenching, and was largely independent of the nature of the particle. P. E. Gibbons and D. C. Northrop (Services Electronics Research Laboratories, Baldock) then described experiments on the long-lived decay components of anthracene scintillations showing that the characteristic times are independent of exciting particles and temperature. These and other experiments on impure anthracene show that these components probably derive from some co-operative lattice property and not a molecular one.

The opening paper of the third session was given by G. Diemer (Philips, Eindhoven), who reviewed present and potential applications of luminescence and photoconductivity in power amplification. The first research paper of the session was read by P. K. Weimer for E. E. Loebner (R.C.A., Princeton) and explained how it should be possible to escape the gain-bandwidth limitations of currently available luminescence amplifiers by using luminor elements themselves capable of modulation and amplification. The second research paper was given by J. Woods (G.E.C., Wembley), who described experiments showing that cadmium sulphide crystals doped with 1–2 per cent copper are *p*-type. Measurements of electrical conductivity, σ, Hall coefficient, R, and thermoelectric power, α, had been made from 95° K. to 380° K., and suggested transport via impurity-levels. At 300° K. it was found for $\sigma = 0 \cdot 18$ ohm^{-1} cm.$^{-1}$ that $p = 1/Re = 5 \cdot 6 \times 10^{14}$ cm.$^{-3}$, $\mu = R\sigma = 20$ cm.^{2} volt^{-1} sec.$^{-1}$, and $\alpha = 22$ μV./°C.

Space-charge-limited current in dielectric crystals formed the topic for the fourth session. The opening paper of this session was given by G. T. Wright (University of Birmingham) and reviewed the mechanisms of space-charge-limited current in dielectrics and, on the basis of a simplified theory and available experimental data, assessed tentatively potential fields of application of space-charge-limited dielectric devices. In a short following paper, A. A. Kayali (University of Birmingham) described measurements of the temperature dependence of forward current of a number of space-charge-limited cadmium sulphide dielectric diodes. In diodes with a small trap density the current increased slowly as the temperature was lowered from 400° K. to 90° K. due to the increase of electron mobility at the lower temperatures; above room temperature there was little variation of characteristics. In most cases the square-law dependence of current upon voltage was obeyed over this whole temperature-range. High-speed switching characteristics of the diodes were described in a paper by A. M. Conning (University of Birmingham). For both 'switch-on' and 'switch-off' operation steady-state conditions were reached in a time less than 3 mμsec., the response time of the measuring equipment; charge storage effects in the diodes were undetectable. The final paper of this session was given by G. T. Wright and discussed the design of a proposed amplifying dielectric triode, which should possess a high input resistance, a large gain-bandwidth product, and be relatively insensitive to temperature changes. Calculations showed that the presence of shallow trapping levels ($< 0 \cdot 30$ eV.) should result in the mutual conductance at high frequencies (> 10 Mc./s.) being many times greater than at audio frequencies.

The opening paper of the fifth session was given by W. J. Merz (R.C.A., Zurich) on the subject of ferro-electricity. Present ideas of basic mechanisms in 'soft' and 'hard' ferro-electrics were reviewed and outstanding problems such as nucleation and losses above the Curie point considered. In the following paper, by K. W. Plessner (British Dielectric Research, London) and R. West (United Insulator Division, T.C.C., Chessington), the influence of the chemical composition of high permittivity ceramics upon their electrical properties was discussed. The application of chemical processes on an industrial scale was then considered. J. H. Bruce and J. R. Balmer (Radio Research Establishment, Malvern) then discussed the need for thin-film capacitors and the suitability of an evaporation method for producing these. Experimental techniques and results for silicon monoxide capacitors were described. The last paper of the session was given by R. C. Kell (G.E.C., Wembley), who discussed piezoelectric ceramics. It had been found that niobate ceramics retained their piezoelectric properties at temperatures considerably higher than ceramics based on barium titanate and that their properties showed less variation with temperature. In particular, sodium–cadmium and lead–barium niobates were more suitable than barium titanate ceramics for use in vibration detectors, ultrasonic generators, and resonators for filter networks.

The final session of the conference included contributions from S. Duinker (Philips, Eindhoven) on a square-loop ferrite device for the fast scanning of electroluminescent cross-bar systems; from D. W. G. Ballentyne (Siemens Ediswan, Harlow) on the possible explanation of electroluminescence as a disorder phenomenon; from R. M. Glaister (G. V. Planar, Ltd., Sunbury) on relaxation polarization dielectrics; from E. A. D. White (G.E.C., Wembley) on the effects of additives on the properties of barium titanate ceramics; from J. C. Burfoot (Queen Mary College, London), who discussed switching in ferro-electrics; from Mr. F. H. Stieltjes (Philips, Eindhoven), who discussed losses in ferro-electrics with particular reference to their use in parametric amplifiers; and G. T. Wright on the trap content of dielectrics with particular reference to the characteristics of dielectric diodes.

The conference emphasized that very great potentialities exist for the exploitation of dielectrics in devices although only limited practical progress has yet been made. There is no doubt, however, that the number and diversity of outstanding problems make this a most attractive field for both fundamental and applied research.

G. T. WRIGHT

THE NATIONAL RESEARCH DEVELOPMENT CORPORATION OF GREAT BRITAIN

IN view of the stress laid on the work of the National Research Development Corporation by Prof. C. F. Carter and Prof. B. R. Williams in "Science in Industry" and of the interest in the Corporation which Lord Hailsham has already shown, the Corporation's annual report for the year July 1, 1958–June 30, 1959, will doubtless be closely examined by scientists and technologists as well as industrialists (National Research Development Corporation. Report and Statement of Accounts for the year 1st July, 1958, to 30th June, 1959. Pp. ii+22. London: H.M. Stationery Office, 1959. 1*s*. 3*d*. net). This year's report is limited to a factual account of the year's work, but the report comments on the greater attention now paid to the Corporation's work by the scientist and industry as well as by the general public, and confidence is expressed that the Corporation's unique experience will enable it to play an ever increasing part in the application of scientific ideas to industry. Revenue from inventions rose to £189,761, and there was also a non-recurring item of £92,978 from sale of patent rights. Government departments and research councils assigned patent rights in 119 cases, compared with 86 in 1958–59, 59 of the total of 185 coming from universities and 5 from industrial research associations, compared with 49 and 1, respectively, out of 147 in 1957–58. Of 681 inventions communicated to the Corporation during the year, compared with 612 the previous year, 287 were from Government departments and research councils, 27 from Commonwealth official organizations, 60 from universities, 8 from industrial research associations and 296 from private firms and individuals, 263 in the United Kingdom. Holdings of British and foreign patent applications totalled 2,794, including 419 United Kingdom patent applications and 580 granted patents, 820 overseas applications and 975 granted patents.

Among new projects initiated during the year or reaching a stage suitable for report are Dr. I. E. Bush's automatic apparatus for chemical treatment and scanning of chromatograms, which has speeded up procedure considerably; the development of the new antibiotics called the cephalosporins, particularly cephalosporin *C*, which is resistant to destruction by penicillinase; Prof. E. C. Cherry's television compression system using a variable scanning velocity so adjusted as to keep the transmitted frequencies within a desired bandwidth. An experimental system based on a 30-line picture has been constructed, and a further experimental system operating on 405 lines is being developed. A contribution has been made towards the cost of equipment to assess a proposal for an improved rolling mill, using rolls supported in hydrostatic bearings, and the Council decided in September 1958 to sponsor the development of Mr. C. S. Cockerell's inventions relating to "Hovercraft", and has formed a subsidiary company, Hovercraft Development, Ltd., to be responsible for its interest in this project. Financial assistance is being provided to complete the development of equipment capable of continuously processing hides and so giving increased capacity allied to smooth-running production and control. As a result of arrangements made by the Corporation to develop and exploit a new type of ditch cleaning media, a British manufacturer is building five prototypes, and the test results of the National Physical Laboratory on the robust and reasonably effective stabilizer designed in 1956 have now been checked on a full-scale ship.

Reference is also made to further developments in a process for producing acetylene by partial combustion of methane, to satisfactory negotiations for financial support of the computer-controlled machine tool project to be taken over by more appropriate organizations ; the completion of a further design study permitting detailed assessment of the economic advantage of Fanshawe's oil-well drilling rig, and of the initial three-year programme of work on the ion-exchange membrane project, under which a process has been developed for continuous electro-chromatography. Considerable further development will be required before the hydrogen–oxygen fuel cell can be launched on a commercial basis. New licensing arrangements were set up with a leading ice-cream manufacturer, who is now in production for the project for a freezing mill after treatment with ultra-sonic waves and pouring into waxed paper containers, and the process appears to offer prospects in the Middle East. Following successful testing and demonstration of the prototype flexible container for transporting oil, seven others have been ordered for use in different parts of the world.

SOLAR STILLS IN IRAQ

By Dr. A. V. BRANCKER
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EXPERIMENTS with solar stills and related units have been in progress at the College of Science, Baghdad, to provide fresh water for Bedouin communities, and to investigate the concentration of natural to heavy water.

Several designs of solar still were tested. Fig. 1 shows a still with a hemispherical 'Perspex' canopy. The base flange was held by screw clips on a gasket in a circular water channel. The complete unit included a flat-plate collector used as a preheater to increase the output of distilled water. The hemispherical still and a prismatic type with a glass canopy have been in continuous operation without maintenance since 1956. Only recently have fine superficial cracks appeared in the 'Perspex'. In other respects these stills, of essentially simple design, have been satisfactory. Curves of water yield versus time and temperature versus time curves for the hemispherical 'Perspex' and prismatic glass stills, with no preheating, are plotted in Fig. 2. The effect of dropwise condensation on the 'Perspex' unit and back reflexion is shown by the relative positions of the curves. The total water output and the dimensions of the stills are given in Table 1.

Table 1

Solar still	Depth of water	Yield of water (litres/sq. m./day)
Hemispherical canopy, radius 1·5 ft.	¾ in.	2·5
Prismatic canopy, 6·0 ft. × 2·2 ft.	¾ in.	3·6

On the basis of 3·60 litres or 0·79 gal. per square metre per day, the output, for a coverage of 1 acre, would be some 3,200 U.K. gal./day. Similar results were obtained by Lof[1] at 0·69–1·07 gal./sq. metre/day. In alternative units, the amount of water from a solar still is approximately 1 lb./sq. ft./day.

In addition to providing fresh-water for small communities, in remote areas, another application is being considered. Thus, natural water contains 1 part of heavy water in 6–7,000 parts ; therefore, very large quantities of water have to be vaporized or processed at heavy fuel costs. The relative volatility of heavy and ordinary water increases with decreasing temperature ; this suggests the possibility of using solar evaporation and fractionation, which can proceed at relatively low temperatures and no fuel costs. One method of investigation is based on the flat-plate solar heater shown in Fig. 1, from

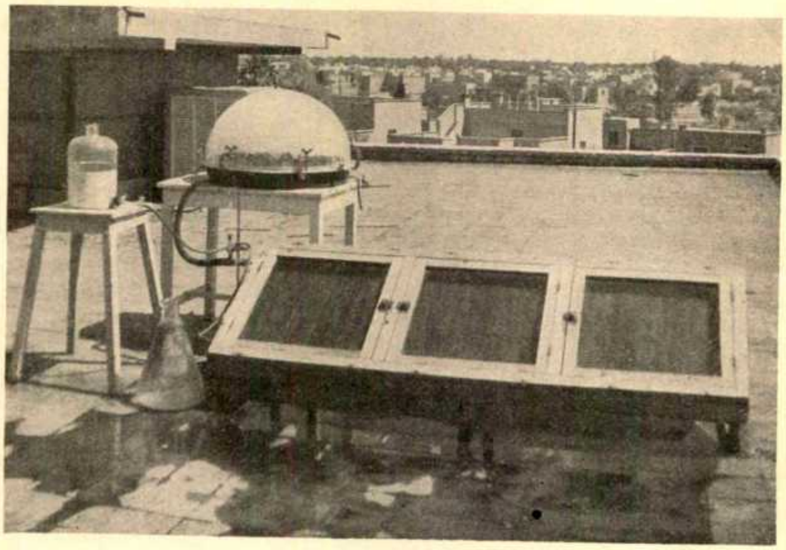
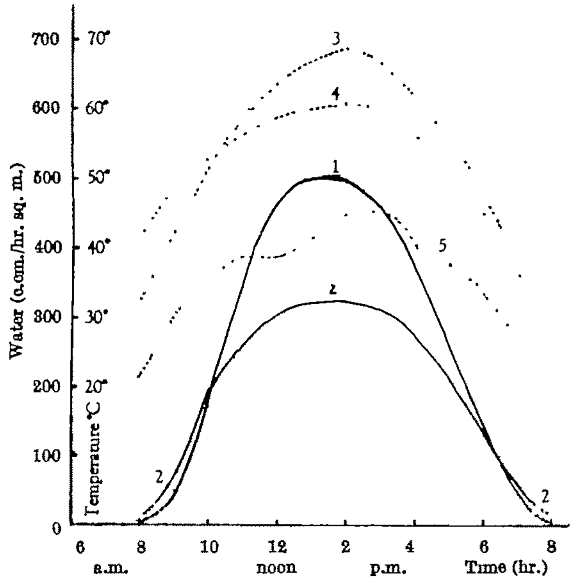


Fig. 1



1, 2, Water yields. 3, 4, 5, Temperatures
1, 3, Glass 2, 4, 'Perspex' 5, Air temp.

Fig. 2

which steam can be obtained. An alternative method is a solar-heated counter-current air–water stripping system. Using a packed tower, water is condensed from hot exit air at saturation humidity. It is not suggested that by solar evaporation methods a full recovery of heavy water is possible, but some preliminary concentration may be obtained for more conventional processing. Again, very large quantities of water would have to be evaporated, but it is precisely this condition which would be necessary in large-scale solar demineralization of brackish water. The objective, therefore, if possible, is to combine two needs.

The physics of evaporation in solar stills is related to the more complex problem encountered in studies of atmospheric evaporation, for which many equations have been derived. It is only possible to outline some methods which can be used. In Table 2, for example, is given a known comparison of heat and mass transfer equations using dimensionless groups.

Table 2

Heat transfer	Mass transfer
(1) Surface conditions	(1) Equimolecular counter-diffusion
$q = h\Delta T$	$N = k_D\Delta P/RT$
$q = -k\,\delta T/\delta x$	$N = -(D_a/RT)\,\delta P/\delta x$
$\theta = T/\Delta T$	$B = P/\Delta P$
$x^1 = x/L$	$x^1 = x/L$
$hL/k = \delta\theta/\delta x^1$	$h_D L/D_a = \delta\beta/\delta x^1$
$\theta = f(x^1, Re, Pr)$	$\beta = f(x^1, Re, Sc)$
$hL/k = f(Re, \nu/\alpha)$	$h_D L/D_a = f(Re, \nu/D_a)$
(2) Energy equation	(2) Continuity equation
$\rho\, DH/Dt - DP/Dt = Jk\nabla^2 T$	$DC/Dt = \mathbf{u}\nabla C + \delta C/\delta t = D_a\nabla^2 C$
$t^1 = vt/L$	$t^1 = vt/L$
$P^1 = P/\rho v^2$	$C^1 = C/\rho$
$H^1 = H/JC_p\Delta T$	$\mathbf{u}^1 = \mathbf{u}/v\ldots.$
$DH^1/Dt^1 - \dfrac{v^2}{JC_p\Delta T}DP^1/Dt^1 = \dfrac{\alpha}{vL}\nabla^2\theta^1$	$\delta C^1/\delta t^1 + \mathbf{u}^1\nabla C^1 = \dfrac{D_a}{vL}\nabla^2 C^1$
Geometrically similar systems	
$qL/k\Delta T = f\left(Re, Pr, \dfrac{v^2}{JC\Delta T}\right)$	$NL/D_a\Delta C = f(Re, Sc)$

The enthalpy balance in B.T.U. for a solar still of area A containing W lb. of brackish water of enthalpy and G lb. of an air–water vapour mixture of enthalpy H_a is given by:

$$WH_w + GH_a = (I - B)At - H_cW_c \qquad (1)$$

where H_c is latent heat of condensation and W_c lb. of water are condensed on the canopy. Assuming $(I - B)$ is constant and differentiating (1):

$$W\mathrm{d}H_w + H_w\mathrm{d}W + G\mathrm{d}H_a = (I - B)At - H_c\mathrm{d}W_c \qquad (2)$$

Further operation on (2) is based on the definitions in Table 3.

Table 3

Enthalpy of water at T_w	$H_w = C_wT_w$
	$\mathrm{d}H_w = C_w\mathrm{d}T_w$
Humidity	$m_a = W_a/G$
	$\mathrm{d}W = \mathrm{d}w_a = G\mathrm{d}m_a$
Enthalpy of air–vapour mixture at T_a	$H_a = C_aT_a + m_a[H_m^{\circ} + C_w(T_a - 32^{\circ})]$
	$= C_aT_a + m_aH_v$
Humid heat	$H_s = C_a + C_w m_a$
Heat transfer	$GH_s\mathrm{d}T_a = h[T_w - T_a]\,A\mathrm{d}t$
Mass transfer	$GH_v\mathrm{d}m_a = k_a\,(P_w - P_a)\,H_vA\mathrm{d}t$
Bowen's ratio for open system (ref. 2)	$R = \dfrac{K}{LE} = 0{\cdot}46\left(\dfrac{T_w - T_a}{P_w - P_a}\right)\dfrac{P}{760}$

K, convection loss to air; L, latent heat evaporation; E, weight of water evaporated.

Applying the definitions in Table 3, it can be shown from equation (2) that:

$$W\Delta H_w + GH_v\mathrm{d}m_a\left[1 + \frac{h}{k_aH_v}\left(\frac{T_w - T_a}{P_w - P_a}\right)\right] = (I - B)At - H_c\Delta W_c \qquad (3)$$

Equation (3), when $\Delta W_c = 0$, may be compared with that for an open system given by Cummings in the nomenclature[3].

$$S + EL\left[1 + 0{\cdot}46\left(\frac{T_w - T_a}{P_w - P_a}\right)\frac{P}{760}\right] = (I - B) \qquad (4)$$

where S is the enthalpy of the water.

Using the heat/mass transfer ratio in Table 3, equation 3 can be simplified to:

$$W\Delta H_w = \left(\frac{M_a}{M_w}k_a P\right)\Delta H_a + (I - B)At - H_c\Delta W_c \qquad (5)$$

where ΔH_w, ΔH_a are the enthalpy changes in time t. From equation 5, if k_a is known, the weight of water evaporated can be estimated. The foregoing and similar enthalpy equations give evaporation-rates of the order obtained experimentally; it is, however, evident from Fig. 2 that equations based on a constant value of $(I - B)$ are only approximations.

The equation for linear conduction in an x-direction is:

$$\mathrm{d}T/\mathrm{d}t = \alpha\,\mathrm{d}^2T/\mathrm{d}x^2 \qquad (6)$$

If the temperature at a surface, $x = 0$, varies in a simple periodic manner:

$$T = T_0\sin\omega t = T_0\sin 2\pi t/P \qquad (7)$$

the solution of equation 6 for boundary condition 7 has the established form:

$$T = T_0\exp(-\mu x)\sin(\omega t - \mu x) \qquad (8)$$

where $\mu = \sqrt{(\omega/2\alpha)}$.

From equation (8), at a depth x, there will be a lag in the temperature wave of phase angle μx; this will amount to a complete period, and phase equality will be attained when $\mu x = 2\pi$ or

$$x = 2\sqrt{(\pi\alpha P)}$$

The solution may be put in the form:

$$\theta = T/T_0 = f(t/P, x/2\sqrt{(\pi\alpha P)}) \qquad (9)$$

which can be applied graphically.

Equations 8 and 9 apply to heat conduction through the Earth or solid material where the heat source is at the surface. In solar evaporation processes there is an absorption of radiation given by:

$$dI_\lambda/dx = -B_\lambda I_\lambda \qquad (10)$$

If the radiation energy is plotted against wavelengths for different depths, the total energy reaching a given depth can be estimated graphically. Evidently the results will depend on the physical and chemical properties of the absorbing medium.

Assuming that:

$$I_\lambda = f(\Delta T, k, \rho, c, L, \lambda, t, \mu) \qquad (11)$$

the results obtained by dimensional analysis are given in Table 4.

Table 4

$$I_\lambda L/\Delta T k = f(\mu c/k, \rho L \lambda/\mu t) \qquad (12)$$

Nusselt number	$N\mu_\lambda = IL/\Delta Tk = (I_\lambda k)/(\Delta T/L)$
Prandtl number	$Pr = \mu c/k = \nu/\alpha$
Modified Reynolds number	$(Re)_\lambda = \rho\lambda L/\mu t = (\rho v L/\mu)(\lambda/vt)$

Geometrically similar systems

$$N\mu_\lambda = f\,[Pr, Re(\lambda/vt)]$$

If, under some conditions, the group:

$$\nu/\alpha \leqslant (\rho v l/\mu)(\lambda/vt)$$

then absorption, at a given wave-length λ at a depth x, is given by:

$$I_{\lambda_0}\{1 - \exp(-B_\lambda x)\}$$

and may depend more on the ratio ρ/μ than on the thermal diffusivity α. Once the radiation is absorbed, and heat generated, conduction will proceed and thermal diffusivity predominate.

These considerations are of interest not only in enthalpy increases and evaporation but also in aqueous quantum processes which occur above a threshold frequency.

The voltage equivalent of the photons is given by:

$$V = hc/e\lambda \simeq 12400/\lambda\text{Å} \qquad (13)$$

Under normal conditions only a small fraction of the total photons may be effective. Also, back reactions reduce the conversion efficiency. If the reaction products could be removed and combined in some ingenious manner, electricity might be generated.

Water, for example, can be decomposed by radiation in the extreme ultra-violet and the electromotive force of the hydrogen–oxygen cell is given by:

$$E = \frac{RT}{zF}\left[\ln K - \ln \frac{C^2_{H_2O}}{C^2_{H_2}C_{O_2}} - \Sigma n_i\right] \qquad (14)$$

When evaporation in a solar still is prevented by a surface layer of inert oil, temperatures approaching 80° C. can be obtained; this may increase the quantum efficiency. In general, for reversible, isothermal processes at constant pressure:

$$zFE = \Delta A_T + \Sigma P_a dV_a = -\Delta G \qquad (15)$$

When $\Delta A_T = 0$ and if $P_1V_1 = P_2V_2$:

$$E = \frac{RT}{zF}\ln\frac{P_2}{P_1} \qquad (16)$$

where P may be a gas pressure or an osmotic pressure in an electrode amalgam.

Further:

$$F\,[E - T(\partial E/\partial T)] = \Delta H \qquad (17)$$

When ΔH (reaction) $= 0$

$$E = \text{constant} \times T \qquad (18)$$

If changes in concentration or pressure can be effected by absorption of solar radiation, the system may function as an engine converting heat into electricity.

In the foregoing and other connexions, solar stills, which can be instrumented for research, offer several interesting possibilities.

Thanks are due to Imperial Chemical Industries, Ltd. (Plastics Division), for the provision of 'Perspex' hemispheres.

[1] Lof, George O. G., U.S. Department Interior, Saline Water Conversion Program, Report No. 5.

[2] Bowen, I. D., *Phys. Rev.*, **27B**, 779 (1926).

[3] Cummings, N., and Richardson, B., *Phys. Rev.*, **30**, 527 (1927). Cummings, N., *Trans. Amer. Geophys. Union*, **21**, 512 (1940); **27**, 81 (1946); **29**, 408 (1948).

MOMENTUM SPECTRUM OF COSMIC RAYS AT SEA-LEVEL

By F. ASHTON, G. BROOKE, M. GARDENER, P. J. HAYMAN, D. G. JONES,
S. KISDNASAMY, J. L. LLOYD, F. E. TAYLOR, R. H. WEST
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PRELIMINARY results are given of the differential momentum spectrum of μ-mesons in the cosmic radiation at Durham (200 ft. above sea-level). The spectrum is used to make an estimate of the production spectrum of the parent π-mesons.

The measurements have been made with the Durham Cosmic-Ray Spectrograph, which consists of a large electromagnet ($\int H dl = 6{\cdot}2 \times 10^5$ gauss-cm.), with layers of Geiger counters and neon flash-tubes as the detecting elements. The overall height of the instrument is 4·95 m. and the aperture is ~ 20 sterad. cm.^2. The design of the instrument is based on the performance of a small prototype spectrograph, described by Ashton *et al.*[1]. The particle trajectories are traced through four measuring-levels, two placed above and two below the magnet. At each level there is a layer of Geiger counters, each counter being attached to an electronic unit giving pulses characteristic of its position in the layer, and eight close layers of neon flash-tubes. The flash-tubes give a precise indication of the position of a particle accepted by the counter selection system.

The measurements at low momenta have been made with the Geiger counter arrangement alone; an electronic momentum analyser computing the deflexion of each particle from the pulses arriving from the counter units at each level. Some 160,000 particles with momenta greater than 1 GeV./c. have been recorded so far. Corrections have been made for the small flux of protons using the proton spectrum measured by Mylroi and Wilson[2], and the resulting μ-meson spectrum is shown in Fig. 1. Only comparative rates are determined and the flux is normalized to that given by Rossi[3] at

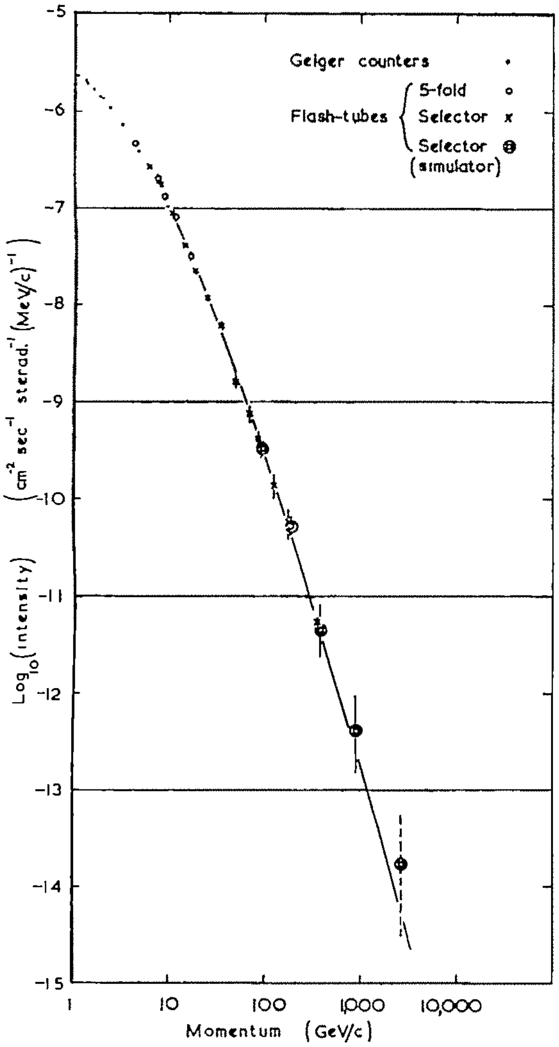

Fig. 1. Differential spectrum of μ-mesons at Durham (200 ft. above sea-level), based on measurements of about 164,000 particles, of which 80 had momenta greater than 100 GeV./c.

1 GeV./c., that is, $2{\cdot}45 \times 10^{-6}$ cm.$^{-2}$ sec.$^{-1}$ sterad.$^{-1}$ (MeV./c.)$^{-1}$.

The measurements have been extended to higher momenta using the arrays of flash-tubes. In order to check the reliability of this technique, a series of photographs has been taken in which the flash-tubes are operated for every particle accepted by the counter system. The measurements are shown in Fig. 1, where they are designated '5-fold' data. The experimental points below 4 GeV./c. have been omitted for clarity. The results are clearly consistent with the counter data.

For good statistical accuracy at higher momenta some selection of particles is required; this is performed by operating the momentum analyser in conjunction with a momentum selector which produces an output pulse only for particles undergoing small deflexions in the spectrograph. Measurements on the flash-tube photographs, made with moderate accuracy using a re-projection system, are sufficient for momenta up to several hundred GeV./c. (designated 'Selector' data in Fig. 1). At still higher momenta a more accurate method is required, and a track simulator is used. With this device a detailed study is made of each track at each level, using the information on track location given by both flashed and unflashed tubes. The maximum detectable momentum of the instrument is (1,000 ± 200) GeV./c. for measurements made in this way; this being the momentum corresponding to a deflexion equal to the probable error of the deflexion measurements. Such results are designated 'Selector (simulator)' in Fig. 1.

Comparison has been made with various other spectra. Below 20 GeV./c. the spectrum is consistent with the results of Owen and Wilson[4] and Caro *et al.*[5]. Between 20 GeV./c. and 200 GeV./c. there is agreement with the spectra of Pine *et al.*[6] and Holmes, Owen, Rodgers and Wilson[7], but not with that of Caro *et al.*[5].

The form of the production spectrum of the π-mesons, whose decay gives rise to the μ-mesons, is an important derivation from the sea-level μ-meson spectrum. Puppi[8] has derived the exponent of the π-meson production spectrum, γ, using the earlier sea-level spectra, with the result that γ is about $2{\cdot}47$ over the range 1–10 GeV./c. and increases slightly at higher energies. Our result is consistent with this conclusion.

Above 20 GeV./c. the exponent has been derived from the analysis of Barrett *et al.*[9]. Using simplifying assumptions, these workers show that a π-meson production spectrum of exponent γ gives rise to a sea-level differential μ-meson spectrum of the form $N(p) \propto \dfrac{1}{p^{\gamma}} \left(\dfrac{1}{1 + p/90}\right)$ (with p in GeV./c.). We have corrected this relation for the momentum loss of μ-mesons in the atmosphere and loss by μ–e decay, and compared it with the experimental results for various values of γ. The exponent giving the best fit is $\gamma = 2{\cdot}55$, and the spectrum using this exponent is shown as the full line in Fig. 1. The conclusion to be drawn from the study of the π-meson production spectrum is that there is little change in the exponent over the range 1–500 GeV./c. This conclusion is of interest in connexion with the high exponent ($\gamma \sim 4{\cdot}3$) inferred from the work on γ-ray cascades reported by Duthie *et al.*[10].

The slope of the sea-level μ-meson spectrum increases continuously with momentum, reaching a value of $-3{\cdot}2$ over the range 100–1,000 GeV./c.

A fuller account of the present work will be published elsewhere.

We thank Prof. G. D. Rochester for his encouragement and interest in all stages of the work.

Grateful acknowledgement is made to the Department of Scientific and Industrial Research for the award of several Special Research Grants and Studentships. Thanks are also due to Profs. P. M. S. Blackett, S. Devons and J. G. Wilson, for their co-operation in making available the large electromagnet.

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[1] Ashton, F., Nash, W. F., and Wolfendale, A. W., *Proc. Roy. Soc.*, A, 253, 163 (1959).

[2] Mylroi, M. G., and Wilson, J. G., *Proc. Phys. Soc.*, A, 64, 404 (1951).

[3] Rossi, B., *Rev. Mod. Phys.*, 20, 537 (1948).

[4] Owen, B. G., and Wilson, J. G., *Proc. Phys. Soc.*, A, 68, 409 (1955).

[5] Caro, D. E., Parry, J. K., and Rathgeber, H., *Aust. J. Sci. Res.*, A, 4, 16 (1951).

[6] Pine, J., Davisson, R. J., and Greisen, K., private communication and Proceedings of the International Cosmic Ray Conference, Moscow, 1959 (in the press).

[7] Rodgers, A. L., Ph.D. thesis, University of Manchester (1956).

[8] Puppi, G., "Progress in Cosmic Ray Physics", 3 (North Holland Pub. Co., Amsterdam, 1956).

[9] Barrett, P. H., Bollinger, L. M., Cocconi, G., Eisenberg, Y., and Greisen, K., *Rev. Mod. Phys.*, 24, 133 (1952).

[10] Duthie, J., Fisher, C., Fowler, P. H., Kaddoura, A., Perkins, D. H., and Pinkau, K., 1960, Proceedings of the International Cosmic Ray Conference, Moscow, 1959 (in the press).

DETERMINATION OF STRONTIUM-90 DISTRIBUTION IN HUMAN BONE

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THE level of strontium-90 in human bone as a result of fall-out has been determined by Kulp *et al.*[1] to be 0·19 μμc./gm. calcium (S.U.) as of January 1958. This value is a world average for adult skeletons. If data for children are included, the average value becomes 0·52 S.U., but there is a variation of the strontium concentration with age, giving a maximum at about 1 year of 1·5 S.U. These levels change with time, however, and Kulp estimates that a maximum concentration in young children will occur at about 1966, on the average corresponding to 4 S.U. and none exceeding 80 S.U. (assuming no further testing).

For an evaluation of the possible biological effects of the strontium burdens cited, it may be of importance to know the distribution of strontium-90 inside the skeleton[2]. From what is known about the metabolism of strontium in bone it is to be expected that the unevenness of the distribution should vary with age and date of birth[3]. For children born after 1954 one would, for example, expect a fairly diffuse and uniform labelling of the bone, while, on the other hand, for individuals born in 1934 or earlier some diffuse and some 'hot-spot' labelling should occur. From, for example, the work of Schulert *et al.*[4] it is evident that an uneven distribution does, in fact, occur if the strontium-90 activity of different bones or different parts of the same bone are compared. The existence at present of hot spots in human bone has, however, not been proved or disproved directly. This would most readily be done with an autoradiographic technique, but the present strontium-90 levels in human bone have been considered too low for this technique to be useful. This assumption will be discussed here. If strontium-90 in bone (varying between 0·1–100 S.U. for different individuals and at different times) is to be studied autoradiographically it is necessary to consider what other activities may be found in bone. For comparison, 1 S.U. corresponds to 0·31 betas/min./gm. of bone from strontium-90 (0·55 MeV.) and the same number from yttrium-90 (2·26 MeV.).

Of the naturally occurring radioisotopes, potassium-40 dominates. 880 p.p.m. potassium in bone[5] gives 1·53 betas/min./gm. bone. Carbon-14 gives 3·2 betas/min./gm. bone[3], but with a maximum energy of only 155 keV. against 1·3 MeV. for potassium-40. Radium-226 is present in small amounts, a body burden of 10^{-10} gm. giving about 0·07 alpha and 0·05 beta/min./gm. bone. Bone may contain other activities besides strontium-90 from fall-out. At present, when no new nuclear weapons tests have been carried out for more than a year, only cæsium-137 could compete. A value of around 0·13 beta/min./gm. bone is to be expected.

If a thick bone sample is autoradiographed, the activities mentioned will have the following importance in approximate relation to 1 S.U. strontium-90: strontium-90, 1·0 by definition; potassium-40, 2·3; radium-226, 1·2; carbon-14, 0·5; and cæsium-137, 0·08. This is comparing the dosages at the surface of a thick bone sample. The maximum ranges of carbon-14 betas and radium-226 alphas are only 100μ and 35μ, respectively, however, and so these contributions may be excluded if a thin absorber is used between the bone sample and the film emulsion. It is still apparent, though, that a study of the present levels of strontium-90 (0·1–2 S.U.) is made difficult by the presence of potassium-40 in bone. This difficulty should, however, be less important in the future if the predicted increase of the strontium-90 levels is found.

In order to avoid disturbances from other activities than strontium-90 and yttrium-90, a new technique of sample preparation has been tried. When a sample of fresh human bone is ashed at a temperature of 700–800° C. it is found that the geometry of the sample changes very little. A section of, for example, the femur, some millimetres thick, becomes slightly twisted and shrunken, but the essential dimensions are almost unchanged. This is true as well for areas of trabecular bone. Using highly active animal bone, we have found that the process of ashing does not change the strontium distribution to any noticeable extent. Hot spots that existed before ashing are still found afterwards and no new concentrations have been created. From what is known about the structure of bone one would expect the geometrical distributions of strontium in fresh and ashed bone to be identical if the bone keeps its geometry during ashing.

The bone sample is white after the ashing, all organic material and particularly carbon (and thus carbon-14) having vanished. The content of potassium-40 has been determined by γ-spectrometry on a 500-gm. femur ash sample. (We are indebted to Mrs. K. Low for making the measurements.) The result was 430 ± 120 p.p.m. potassium in bone ash, which corresponds to 180 ± 50 p.p.m. in fresh bone. This value is only 20 per cent of that in ref. 5. The difference may perhaps be explained by variations in the preparation of the bone before ashing. In our case, as much as possible of the organic material was removed beforehand. The sample measured was too small to demonstrate the presence of any cæsium-137.

The low value found for potassium-40 in bone ash reduces the effect on the determination of strontium-90 discussed previously from 2·3 to less than 0·5 (strontium-90 = 1·0). A still further reduction may be achieved if the ashed bone sample is carefully soaked and washed with water or ammonium carbonate solution. In this way, 570 p.p.m. of potassium-40, that is, between 90 and 100 per cent, has been extracted from the above-mentioned femur sample. The same is most probably true for cæsium-137 as well, although it has not been possible to check this as yet. A similar extraction carried out on highly active (strontium-90) animal bone revealed that only a small fraction (1–2 per cent) of the strontium is removed in the process. This should apply to radium-226 as well. It is thus possible to prepare bone samples which contain almost only strontium-90 + yttrium-90 and radium-226 activity (for 1 S.U.: strontium-90 = 1·0; radium-226 ~ 1·2; potassium-40 < 0·05; cæsium-137 < 0·01; and carbon-14 ~ 0).

One might now ask what times of exposure are necessary to produce an autoradiograph of these low levels of strontium-90. 1 S.U. corresponds to about 1 beta/cm.^{2}/hr. on the surface of a sample of bone[6]. Using X-ray film ('Kodirex') it has been reported[7] that a good autoradiogram is produced in 80 days with a bone sample containing not more and probably less than 126 S.U. In about the same time 'a very slight image' was produced with a sample containing not more than 11 S.U. Similar results have been obtained in our laboratory. These exposures should correspond to about 250,000 and 25,000 betas/cm.^{2}, respectively. It is interesting to note that the hard cosmic-ray component would in an 80-day exposure contribute about 120,000 tracks/cm.^{2} of emulsion. To make possible autoradiographs on the 1 S.U. level this high background must be reduced. A factor of 10 may, however, be gained through storage in a shelter with a 20-m. rock roof. Additional lead shielding of the films is then required to absorb the γ-radiation from both the rock itself and from radon with daughter products. In this way, a film background over 80 days of less than 10,000 tracks/cm.^{2} should be a possibility.

In addition to this it would be necessary to reduce the thermal and chemical blackening that takes place during storage and which over a year may increase from, for example, 0·06 to 0·30 (ref. 8). A twofold reduction of this blackening results if the film is stored at low temperature (between − 5° C. and + 5° C.). The blackening due to carbon-14 and tritium in the emulsion, finally, is negligible in comparison with the cosmic ray background.

Granted that underground cold-storage is effective in reducing the background blackening of X-ray films, it is reasonable that 100,000 betas/cm.^{2} should produce a good autoradiogram and 10,000 betas/cm.^{2} a very slight one on 'Kodirex' film. For 1 S.U. this corresponds to exposure times from about 10 and 1 years, respectively. Comparing 'Kodirex' with other X-ray films, the following figures for relative speed may be cited[8]: 'Kodirex', 35; '*CEA* Textur-*X*', 47; 'Ilfex', 52; Ilford Industrial *G*, 57; Kodak Industrial *K*, 77; and Fuji Industrial 200, 100. Up to a factor of 2–3 in exposure time may thus be saved using films that have high speed. Additional time may possibly be saved if local strontium-90 concentrations occur.

A second way to use autoradiography to study the labelling of human bone with strontium-90 is with β-sensitive nuclear emulsions. To begin with, the emulsion would have to be cleaned from the existing background of β-tracks. This is possible with a water-vapour treatment for some hours at 50–60° C.[10]. Subsequent exposure to attain 10^3–10^4 betas/cm.^{2} may produce results that are measurable by microscopic scanning[11].

It may thus be possible, using an exposure time of one year, to get a weak autoradiogram of bone carrying 1 S.U. diffusely distributed. This type of distribution corresponds to that of bones from children younger than fifteen years (0·7–1·5 S.U.). For adults having an average of 0·19 S.U. (January 1958) it is to be expected that local concentrations will occur, some parts of the bone containing strontium-90 in excess of 1 S.U. This should again be observable with exposure times of about a year. An increase of the future strontium-levels would make the autoradiographic method still more feasible.

[1] Kulp, J. L., Schulert, A. R., and Hodges, E. J., *Science*, 129, 1249 (1959).

[2] Björnerstedt, R., and Engström, A., *Science*, 129, 327 (1959).

[3] Report of the United Nations Scientific Committee on the Effects of Atomic Radiation, Supp. 17 (*A*/3838) (1958).

[4] Schulert, A. R., Hodges, E. J., Lenkoff, E. S., and Kulp, J. L., *Health Phys.*, 2, 62 (1959).

[5] Report of the Internal Dose Committees of the I.C.R.P. and N.C.R.P.-1958 Revision, B. Lindell (private communication).

[6] Rossi, H. H., and Ellis, jun., R. H., *Nucleonics*, 7, No. 1, 18; No. 2, 19 (1950).

[7] Hawthorn, J., and Duckworth, R. B., *Nature*, 182, 1294 (1958).

[8] Sjöstedt, G., CEA-verken, Strängnäs (private communication).

[9] Smits, D. W., *et al.*, *Acta Cryst.*, 9, 520 (1956).

[10] Marguin, G., Wissenschaftliche Photographie, Internationale Konferenz, Koln (1956).

[11] Kristiansson, Dr. K., Lund (private communication).

POTATO WART DISEASE, *SYNCHYTRIUM ENDOBIOTICUM* (SCHILB.) PERC.

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IT was thought in the past that the potato wart fungus could exist equally well in cold as in warm or even sub-tropical regions, that is, in all places where potatoes[1,2] are cultivated. This opinion diverted attention from the investigation of ecological factors affecting the fungus in its environment. Uncertainty about this led me to a detailed study of the genesis and disappearance of the first occurrence of potato wart disease in Europe[3,4], to the laying down of field tests in sub-arid and arid regions, and to the utilization of data about the historical and present occurrence of potato wart disease throughout Europe.

Analysis of the climatic conditions of the district of Hornany in Czechoslovakia, where potato wart disease was first described[4], shows that the disease ceased to exist after a drier and warmer period[3].

Field tests in the sub-arid region of Czechoslovakia showed that among tubers harvested in humid regions from land in an area where there had been a marked appearance of potato wart disease no case of the disease was found. However, when every potato plant was given 5 kgm. of totally infected soil from the humid area where the Wohltmann variety crop was 90–95 per cent destroyed, a small amount of wart disease was registered in the first year, a smaller amount in the second year, and little or nothing in the third year (Fig. 1). Very often the out-growths were atypical. When in the period 1951–58 the gradually infected soil was brought back from the sub-arid region to a humid area (5 kgm. of totally infected soil for each potato plant) the appearance of wart disease was detected as shown in Fig. 2. It was also found that when totally infected soil from the humid area was placed in the sub-arid region without being mixed with the soil of that region the amount of wart disease was greater, and the cleansing of the soil from the fungus slower. In the fourth year the disease was sporadic or lacking. In the humid area in the same soil wart disease was proportionately greater (Fig. 3).

Judged from the results obtained, the practical eradication of the potato wart fungus from the soil in sub-arid regions does not take long even when the soil is strongly infected (2–3 years), though complete eradication takes longer (5–7 years) and this happens even when non-immune varieties of potatoes are grown.

Analysis of the occurrence of wart disease in European States from the climatological–geographical point of view demonstrated that the greatest intensity is in the mountainous and sub-mountainous regions of central, northern and eastern Europe. In the maritime countries by the North Sea (Great Britain, Benelux, Northern France) there is a smaller and economically less significant amount, however humid and mild the climate. In the warmer regions of southern Europe, and the sub-arid plains of central Europe having a continental climate, wart disease has not been reported.

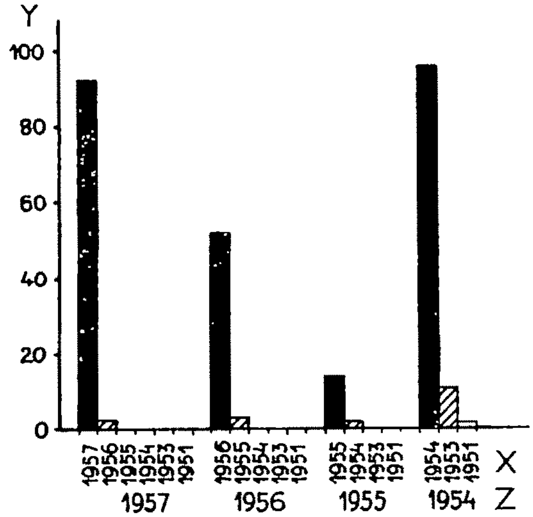

Fig. 1. Intensity of wart disease in sub-arid regions on soil highly infected (5 kgm. totally infected soil from the humid area to each potato plant). *X* = year of infestation of the soil; *Y* = weight of outgrowths in gm.; *Z* = year of the experiment

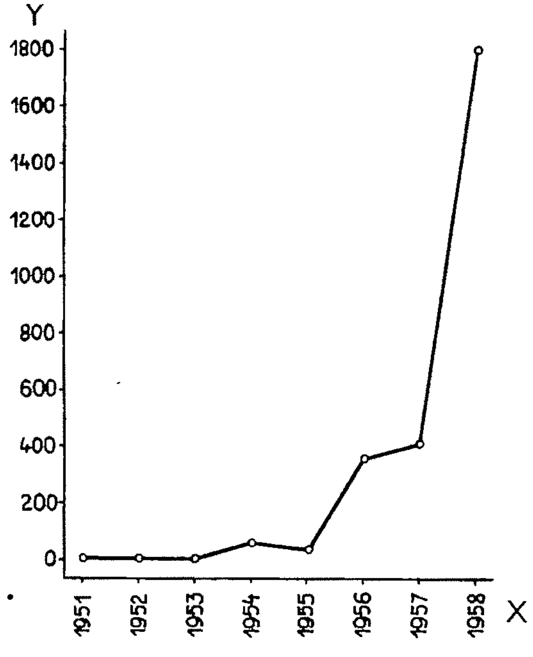

Fig. 2. Intensity of wart disease in the soil gradually infested (5 kgm. totally infested soil to one potato plant) in the sub-arid region in 1951–58 and in 1958 brought into humid region. *X* = year of infestation of the soil; *Y* = weight of outgrowths in gm.

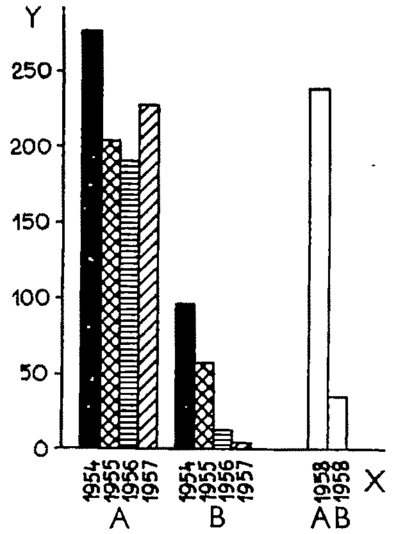

Fig. 3. Intensity of wart disease in totally infected soil, placed in 1954 in the humid *A* and sub-arid *B* regions and in 1958 returned to the humid region. *X* = year of experiment, *Y* = weight of outgrowths in gm.

Temperature and rainfall, and to a certain extent the nature of the soil, are the most decisive factors in the successful or unsuccessful development of the fungus.

The potato wart fungus likes climates which may be characterized thus: low summer temperature (under 18° C.), long and deep winters (160 and more days with temperatures below 5° C.), heavy precipitation (700–800 mm. and more per annum, the greater part during the summer), sufficiently aerated and extensively cultivated soil, with an acid reaction and poor rotation of crops, where potatoes are planted every second or third year. In areas with mild, short winters many zoosporangia germinate without infection taking place. Rapid cleaning of the soil follows, and the appearance of wart disease is sporadic or nil and economically of no importance.

The results of the investigations have made it possible, first, to establish for Europe a prognosis of the occurrence of potato wart disease, according to which suitable, less suitable, and unsuitable zones can be fixed for the contamination and development of potato wart disease; secondly, to regulate the breeding and cultivation of potatoes. In the unsuitable zone it is possible without apprehension to cultivate non-immune varieties of potatoes, many of which are more tolerant to degeneration and important from the point of view of production of early potatoes. Finally, it is possible to solve plant quarantine questions, and control the disease according to the individual ecological–geographical zones.

Further details will be given in my book, "Ecology and Prognosis of Potato Wart Disease", to be published with an extensive English summary by the Slovak Academy of Sciences Press in 1960.

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[1] Bordukova, M. B., Bolezni i vrediteli kartofl'a. Moskva, Gos. Izdat. selchoz. lit. (1955).

[2] E.P P.O., Potato Wart Disease in Europe in 1954: European and Mediterranean Plant Protection Organization, Paris (1955).

[3] Bojnanský, V., *NachrBl. deutsch. PflSchDienst, Berl.*, 11, 109 (1957).

[4] Schilberszky, K., *Ber. deutsch. bot. Ges.*, 14, 36 (1896).

LETTERS TO THE EDITORS

PHYSICS

Self-quenching Geiger Counters containing Mixtures of Permanent Gases

We have investigated the characteristics of Geiger–Müller counters containing mixtures of argon with small quantities of xenon (0–7 per cent), oxygen (0–0·5 per cent) and nitrogen (~ 1 per cent), to total pressures of 700 mm. mercury. That counters filled with this mixture would behave as 'fast' counters in a manner similar to organically quenched counters was first reported by Shore[1], although Riedel[2] reported that using the filling recommended by Shore produced no Geiger region. Using counters of the conventional 'gamma' type, glass bodies with cylindrical cathodes of various materials together with a 4·7 megohm series resistor and a linear amplifier of a gain of about 25, we find that optimum proportions of the gas mixture to produce the best plateaux are: xenon (3·5 per cent), oxygen (0·3 per cent), nitrogen (1·2 per cent), with argon added to give a total pressure of 700 mm. mercury. Such a mixture gave a plateau about 170 V. in length, with a starting voltage of 1,460 V. and a plateau slope of 0·02 per cent/V.

Some fairly general conclusions can be reached as to the role of the various components in the mixture in producing the Geiger plateau. It is evident that the oxygen makes possible the rapid spread of the discharge along the counter anode, chiefly by photoionization, in the manner described by Huber[3]. This was made clear when fillings of pure argon and argon plus 1 per cent xenon were investigated, by examination of the discharge by oscilloscope. It was found that with both these mixtures there was no Geiger region, the discharge passing abruptly from the proportional to the corona region, the voltage at which this occurred being lowered from 1,850 V. to 1,260 V. with the addition of xenon. Addition of 0·1 per cent of oxygen to pure argon altered the character of the discharge entirely, the familiar Geiger pulses being observed although a plateau of only a few volts was obtained with many spurious pulses.

Although Shore reported that, if nitrogen were not present in the mixture, the plateau became extremely short, we have not found this to be so. The addition of about 1 per cent of nitrogen improves the performance of the counters but is not essential. For example, a counter containing the optimum quantities of xenon and oxygen had a plateau of 102 V. in length with a slope of 0 064 per cent/V. Refilling and adding 1 per cent of nitrogen resulted in an increase in length to 130 V. and the slope improved to 0·025 per cent/V.

It is also clear that the presence of the xenon inhibits the formation of the many spurious pulses found in the argon–oxygen–nitrogen mixtures. Adding small quantities of xenon to the latter results in an improvement in length and slope of the plateau together with a very sharp fall in the starting voltage, from 2,124 V. for zero quantity to 1,460 V. for the optimum amount of xenon (3·5 per cent). Adding further xenon results in an increase in the starting voltage, and a deterioration of the slopes and lengths of plateaux.

In an endeavour to understand this mechanism we have investigated the time incidence of spurious pulses by pulse-interval analysis[4] for mixtures containing increasing quantities of xenon with fixed amounts of nitrogen (1 per cent) and oxygen (0·3 per cent). At the same time we have measured the transit times of the front of the positive-ion sheath from examination of the cathode-current pulse. In all the many fillings investigated we have found that the maximum number of spurious pulses occur at the end of the counter dead-time and only a negligible proportion are recorded after the front of the positive-ion sheath has reached the cathode. It is apparent, at any rate with these counters, that the positive-ion effect at the cathode, often quoted as the reason for the generation of spurious pulses in Geiger counters, is unimportant.

The other possible mechanism is that the spurious pulses are due to hard photons, delayed relative to the main Geiger pulse, releasing photo-electrons at the cathode. Such hard photons could arise as a result of the quenching of argon metastables by collision with gas molecules. In fact, pulse-interval analysis of mixtures containing zero or less than optimum quantities of xenon shows large numbers of small spurious pulses occurring in as short a time as 15 μsec. after the main Geiger discharge. With optimum quantities of xenon such small pulses do not appear and the counters have a clear dead-time of about 150 μsec, indicating that the argon metastable states are rapidly quenched by collisions with xenon atoms. That this occurs, and with the formation of ions, is also indicated from the considerable lowering of starting voltage with addition of xenon, while the charge per pulse remains of the same order. The ionization potential of xenon (12·127 V.) is higher than both metastable levels of argon (11·53, 11·72 V.) so that direct ionization of xenon atoms by collision with argon metastable states is not possible; but in spite of this it seems fairly evident that some mechanism exists involving interaction between xenon atoms and argon metastable states giving rise to an increase in the number of ions produced in the avalanches.

A more detailed report of the work described in this communication is to be published elsewhere.

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[1] Shore, L. G., *Rev. Sci. Instr.*, **20**, 956 (1949).

[2] Riedel, O., *Z. Naturforsch.*, **5a**, 331 (1950).

[3] Huber, E. L., *Phys. Rev.*, **97**, 261 (1955).

[4] Putman, J. L., *Proc. Phys. Soc*, **61** 312 (1948).

An Image Intensifier with Transmitted Secondary Electron Multiplication

THE use of transmitted secondary electron emission for electron multiplication has been proposed by Lubszynski[1], McGee[2] and Sternglass[3], and photoemissive image intensifiers incorporating thin films of potassium chloride as transmitted secondary emitters have been described by Wachtel, Doughty and Anderson[4]. We also have been experimenting with the construction of image intensifiers of this kind.

The form of the tubes we have made is shown in Fig. 1. They consist essentially of an antimony–cæsium photocathode, an aluminium-backed zinc sulphide–silver phosphor, and between these a set of parallel electron-multiplying dynodes which are layers of aluminium and potassium chloride deposited on a supporting film of aluminium oxide. Electrons from the photocathode are accelerated and focused on the first dynode by means of coaxial electric and magnetic fields, and secondary electrons are produced which escape from the dynode on the side remote from the photocathode. These transmitted secondaries are similarly accelerated and focused on the second dynode, where further transmitted secondary electrons are produced; and this process is repeated down the tube. Finally, the electrons which emerge from the last dynode are accelerated and focused on the phosphor.

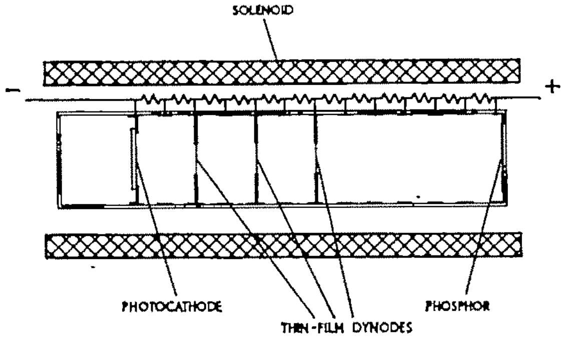

Fig. 1. Image intensifier with transmitted secondary electron multiplication

The electric field is provided by cylindrical electrodes on the inner wall of the tube, which are connected to an external potential divider. The magnetic field is provided by a long solenoid which surrounds the tube. In order to prepare the photocathode without admitting cæsium to the working section of the tube we have made use of the arrangement described by McGee[5]. At a short distance from the front window of the tube there is an annular metal shelf which fits the inside of the tube closely enough to act as a barrier to cæsium vapour, and a glass plate is held to this shelf by means of a spring catch which can be released by tapping the outside of the tube. The photocathode is formed on the surface of the plate which faces the front window of the tube. After the tube is sealed off from the pumps, the plate is released and turned over so that the photocathode faces the dynode system.

The tube we have prepared most recently has five 1-cm. diameter dynodes, each consisting of a layer of aluminium ~200 A. thick and a layer of potassium chloride ~500 A. thick deposited on a film of aluminium oxide ~450 A. thick. The sensitivity of the photocathode is 20 μamp./lumen. We have operated this tube with 37 kV. between photocathode and phosphor. The potential difference between the cathode and the first dynode, and between successive dynodes, is then about 4·9 kV., and the magnetic field required for focus is 180 gauss. Under these conditions the total electron multiplication is $\sim 3\cdot5\times10^{3}$, and the ratio of total light output to light input is $\sim 8\times10^{4}$ when the incident light is of the same mean wave-length as the phosphor emission. The sensitivity of the photocathode corresponds to a quantum efficiency of about 4 per cent at this wave-length, so that each electron leaving the cathode must result on average in a pulse of about 2×10^{6} photons from the phosphor. These pulses are easily visible

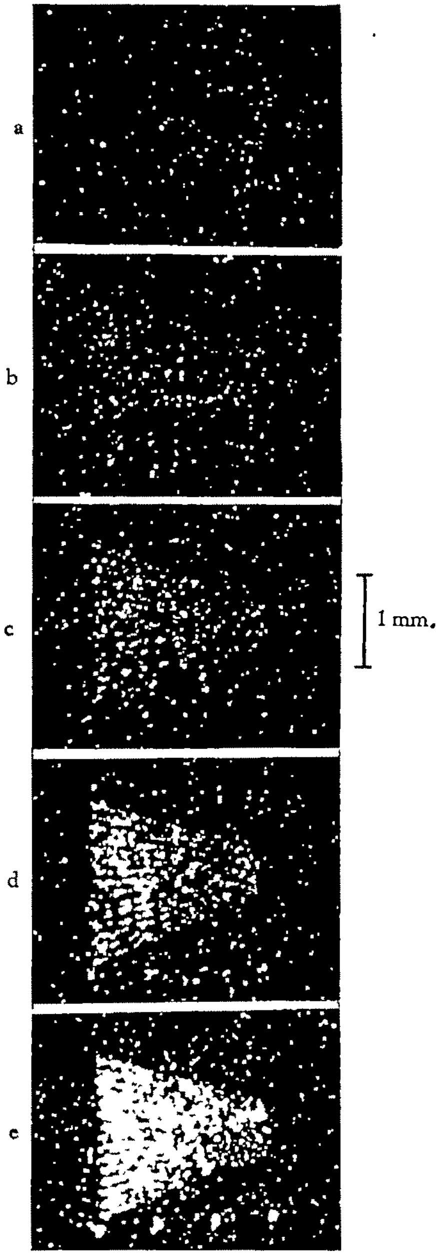

Fig. 2. Photographs obtained with image intensifier showing scintillations resulting from single electrons leaving the photocathode: *a*, dark background; *b*, *c*, *d*, *e*, image of fan-shaped test object projected on photocathode with successively increased levels of illumination. Exposure times 0·5 sec.

with a low-power microscope as bright scintillations on the phosphor, with diameters of about 40 microns. For an average scintillation the photon density at the phosphor thus exceeds 10^{11} photons cm.$^{-2}$, and since a photon density of 10^{8} photons cm.$^{-2}$ is sufficient to give a detectable image on a very fast photographic emulsion[6], it is to be expected that the scintillations can be photographed with relatively inefficient optical coupling between the phosphor and the emulsion.

We have been able to take such photographs (Fig. 2) on Kodak Tri-*X* film, using a pair of 50 mm. *f*/2 lenses front-to-front to give unity magnification. The exposure time for each photograph was 0·5 sec. Fig. *2a* was taken with the tube in the dark at room temperature, and shows the scintillations due to dark emission from the photocathode. The rate of background scintillation is $\sim 6 \times 10^{3}$ cm.$^{-2}$ sec.$^{-1}$. The corresponding cathode dark current, if each electron emitted gives rise to a scintillation, is $\sim 10^{-15}$ amp. cm.$^{-2}$, which is a reasonable value for an antimony–cæsium cathode. For Figs. *2b*, *c*, *d* and *e*, the image of a resolution test object was projected on to the photocathode with successively increased levels of illumination. The test object was a fan of 15 black-and-white line-pairs. In Fig. *2b* the illumination is so low that the photo-emission is only of the same order as the dark emission, and the object is detectable only as a local increase in the scintillation density. Increases in illumination lead to the object becoming barely recognizable (Fig. *2c*), and clearly so (Figs. *2d*, *e*). The limiting resolution is about 15 line-pairs per mm., and is determined largely by the diameters of the scintillations.

We are grateful to Prof. J. D. McGee for his interest and encouragement, and to the Department of Scientific and Industrial Research for financial support.

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[1] Lubszynski, H. G., Brit. Pat. No. 457493 (1935).

[2] McGee, J. D., Brit. Pat. No. 504927 (1937).

[3] Sternglass, E. J., *Rev. Sci. Instr.*, 26, 1202 (1955).

[4] Wachtel, M. M., Doughty, D. D., and Anderson, A. E., Proc. Symposium: Photo-electronic Image Devices ("Advances in Electronics", 12) (Academic Press, New York and London; in the press).

[5] McGee, J. D., Proc. Symposium: "The Present and Future of the Telescope of Moderate Size", 35 (Univ. of Pennsylvania Press, 1956).

[6] Value quoted by Perl, M. L., and Jones, L. W., Proc. Symposium: Photo-electronic Image Devices ("Advances in Electronics", 12) (Academic Press, New York and London, in the press).

Ultrasonic Velocities in Aqueous Solutions of Cadmium Iodide and Mercuric Chloride

In the course of a systematic study of the ultrasonic behaviour of aqueous solutions of inorganic salts, I have come across two cases (mercuric chloride and cadmium iodide) which behave in a peculiar manner. Preliminary measurements on the velocity of ultrasonic waves in aqueous solutions of the above salts have been made using a variable path interferometer working at 750 kc./sec. An accuracy of 0·15 per cent is obtained. The results are presented graphically in Fig. 1.

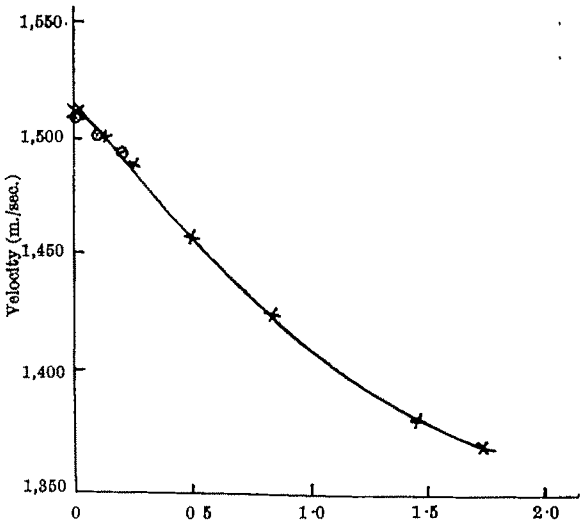

Fig. 1. ×, cadmium iodide; ○, mercuric chloride

In contrast with the normal behaviour of electrolytes, ultrasonic velocities of these solutions decrease with increasing concentration. Cadmium iodide registers a large decrease. Ultrasonic velocity at a concentration of 1·75 *M* is 1,369 m./sec., resulting in a lowering of 140 m./sec. from the value for pure water. Mercuric chloride shows a relatively smaller change, the velocity in its 0·25 *M* solution being 1,489 m./sec.—a lowering of 18 m./sec. Even though the ultrasonic velocities decrease with increasing concentration, the adiabatic compressibility decreases with increase of concentration. Full details of this investigation will be published shortly.

I am indebted to Dr. J. Bhimasenachar for his encouragement and helpful discussions during the course of this work.

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A Sidewall Friction Effect during Soil Consolidation

In the course of investigating the consolidation properties of a sensitive marine clay, a variety of consolidation test procedures has been used. Some followed normal incremental loading procedure, but with widely varying increment ratios. Other tests employed continuous loading techniques either by straining the specimen at a constant rate or by applying load at a constant rate.

When the specimens were oven dried after test, an unusual phenomenon was observed. Specimens that had been subjected to incremental loading revealed a dark ring of soil at mid-height around the perimeter. During the drying this ring of soil appeared to shrink more than the rest of the specimen; it often fell away leaving a spool-shaped specimen as shown in Fig. 1. The phenomenon was never observed on specimens subjected to continuous loading.

In a paper[1] which describes this work more fully, it is reasoned that the differential shrinkage in specimens loaded in increments is a manifestation of unequal stress distribution across the specimen and that sidewall friction between specimen and enclosing ring is the primary cause of the non-uniformity. Reference to probable effective stresses through incrementally loaded specimens suggests that unequal consolidation will occur, but that in continuous

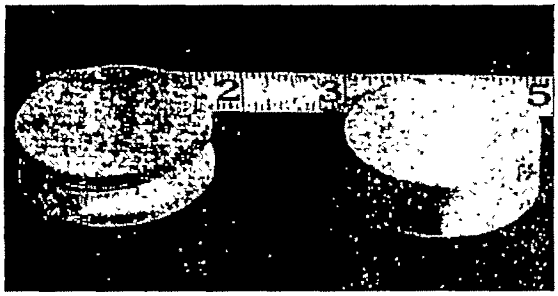

Fig. 1. Differential shrinkage of oven-dried sample due to unequal stress distribution. Specimen on left subjected to incremental loading $\Delta p/p = 1$; specimen on right tested by a continuous load increase technique

loading tests (pore water pressure assumed to be zero) a more uniform consolidation is visualized. This appears to have an important effect on the measured preconsolidation pressure. Although the phenomenon reported was observed in a highly compressible, sensitive clay it may be found in other soils.

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[1] Hamilton, J. J., and Crawford, C. B., "Improved Determination of Preconsolidation Pressure of a Sensitive Clay", submitted to the Amer. Soc. Testing Mat.

METALLURGY

Interstitials and Fracture of Metals

INTERSTITIAL alloying elements, even in minute quantities, exert a potent influence on the mechanical behaviour of alloys. Of particular recent interest has been the behaviour of hydrogen in steels; where the phenomenon of hydrogen-induced delayed brittle failure has received special attention[1–3]. Time-dependent brittle failure implies that spontaneous brittle fracture will occur at loads substantially below those that may have been previously successfully sustained. The characteristic features are: (*a*) an incubation period; (*b*) slow crack propagation; (*c*) a lower critical stress below which failure will not occur. Both the incubation period for crack initiation and crack growth are controlled and paced by the diffusion of hydrogen[4–6,8].

The incubation period depends on the time required to accumulate a critical concentration of hydrogen in the region of maximum triaxiality. Diffusion to this region is induced by the strain gradient associated with the inhomogeneous stress field, wherein the strain gradient acts as an activity gradient or driving force. It has been demonstrated recently[9] that the attainment of the critical hydrogen concentration is determined by the magnitude of the applied stress. That is, the applied stress governs the magnitude of the strain gradient. High triaxial stress states are brought about by notches. The notches may be artificial (excavated), inherent (inclusions, grain boundary intersections, dislocation arrays, etc.) or combinations of these. However, when brittle fracture is involved, modern theory requires one of the effective notches to be a dislocation array. Therefore, coupled with the strain gradient of an artificial notch may be an additional driving force associated with a dislocation array at the base of the notch. All this suggests that there are two criteria for delayed brittle failure; specifically, a critical concentration of hydrogen and a critical stress state. These two criteria are the basis of the discontinuous nature of crack propagation which is a unique characteristic of this type of time-dependent brittle failure.

The occurrence of discontinuous crack propagation makes it difficult to rationalize delayed brittle failure by applying any mechanism which postulates raising the flow stress to the fracture strength. On the other hand, a decrease in the cohesive strength of the lattice provides a ready explanation for this phenomenon. This could be brought about by localized concentration of interstitial atoms in the region of the critical stress state in the form of an uncondensed atmosphere[10]. Probably a more general situation would be one which involves both a rise in flow stress brought about by plastic deformation and a lowering of the cohesive strength. Presumably the proportions of these two could vary over a wide range.

Once the crack has been initiated, it will be propagated until it exceeds the radius of influence of the accumulated interstitials. Beyond this point the moving crack will be damped out rapidly due to plastic flow ahead of the crack. Another incubation period is then required to build up the critical concentration at the tip of the crack. This step-like process continues until the effective area is reduced such that the applied stress is equal to the notched strength of the material. Thus, a lion's share of the time to failure resides in the sum of the successive incubation periods.

No restrictions are apparent which limit this behaviour to hydrogen and steel. Indeed, the only restrictions to a general behaviour of this type are that a critical stress state exists and that the interstitials have sufficient mobility to attain a critical concentration in a reasonable length of time. Hence, one may well anticipate that any interstitial in any metal which can accommodate the critical stress state may induce delayed brittle failure.

A major step in evaluating this concept involved an examination of the particular case of carbon (nitrogen) in steel. In the temperature-range 700–1,000° F. carbon has approximately the same mobility

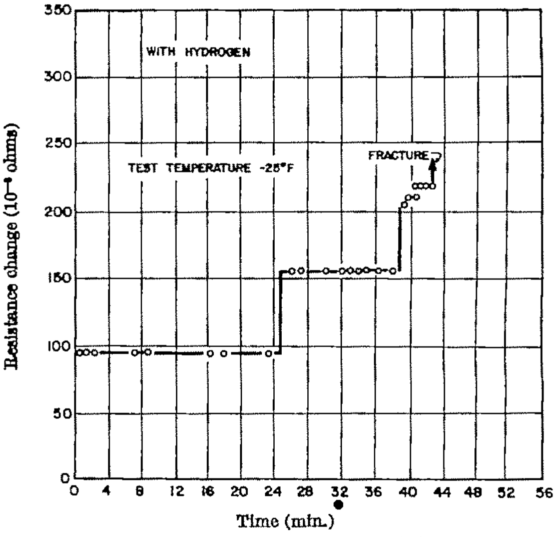

Fig. 1. Resistance vs. time in the delayed-failure stress range of a uniformly hydrogenated nickel–chromium–molybdenum steel

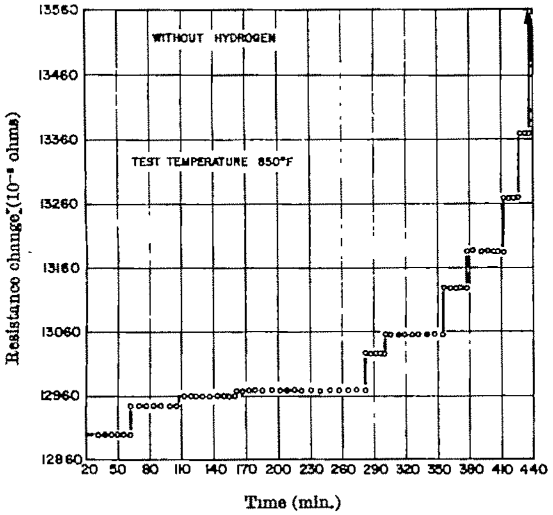

Fig. 2. Resistance vs. time in the delayed-failure stress range of a high-temperature die steel

in steel as has hydrogen at room temperature. Notched specimens of a high-temperature die steel (0·40 per cent carbon, 0·35 per cent manganese, 1·10 per cent silicon, 5·00 per cent chromium, 0·90 per cent vanadium, 1·35 per cent molybdenum) were subject to sustained-load-to-failure tests in the appropriate temperature-range. Resistance versus time measurements[3,4] were recorded during the sustained-load tests. All the significant features of time-dependent brittle failure were observed in an almost perfect image of the behaviour of hydrogen in steel (cf. Figs. 1 and 2). Fig. 2 vividly illustrates the incubation period and the step-like appearance associated with discontinuous crack propagation.

These results lend authority to the concept that interstitial-induced delayed brittle failure may be a general behaviour in metals which can attain the critical stress state in the temperature-range dictated by the mobility requirement. The implications derived from this work are far-reaching. They indicate that 'pressure in a void' is unnecessary to explain time-dependent brittle failure. In addition, they suggest the the possibility of unifying this behaviour with other, more familiar, phenomena which also are paced by the diffusion of interstitials, such as slow-strain-rate embrittlement and strain ageing[10].

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[1] Petch, N. J., and Stables, P., *Nature*, **169**, 842 (1952).

[2] Frohmberg, R. P., Barnett, W. J., and Troiano, A. R., *Trans. Amer. Soc. Met.*, **47**, 892 (1955).

[3] Barnett, W. J., and Troiano, A. R., *Trans. Amer. Inst. Mining and Met. Eng.*, **209**, 486 (1957).

[4] Johnson, H. H., and Troiano, A. R., *Nature*, **179**, 777 (1957).

[5] Johnson, H. H., Morlet, J. G., and Troiano, A. R., *Trans. Amer. Inst. Mining and Met. Eng.*, **212**, 528 (1953).

[6] Troiano, A. R., *Corrosion*, **15**, 207 (1959).

[7] Klier, E. P., Muvdi, B. B., and Sachs, G., *J. Metals*, **9**, 106 (1957).

[8] Steigerwald, E. A., Schaller, F. W., and Troiano, A. R., *Trans. Amer. Inst. Mining and Met. Eng.*, **215**, 1048 (1959).

[9] Steigerwald, E. A., Schaller, F. W., and Troiano, A. R. (to be published).

[10] Troiano, A. R., 1959 Campbell Memorial Lecture, Amer. Soc. Met. (to be published).

Determination of Oxygen in Iron and Steel

WE have recently applied the method of stable isotope dilution[1] to the determination of oxygen in samples of iron and steel which contained low concentrations of this element. As our results are consistently higher than values obtained by the conventional vacuum-fusion method, this preliminary account of our work is submitted.

The technique consists of allowing the steel sample to react with carbon at 1,500° C. in an atmosphere containing a known amount of carbon monoxide enriched in the isotope oxygen-18. Measurement of the ratio $R = {}^{18}O/{}^{16}O$ in a sample of the gas after equilibration enables the oxygen content of the metal to be determined. 'Blank' corrections are negligible, but as an additional precaution may be avoided by prior equilibration of the graphite crucible used as container for the sample in an atmosphere containing the same relative amounts of oxygen-18 and oxygen-16 as the expected final equilibrium value R.

Various methods have been used to determine the ratio R. At first, use was made of the mass spectrometer ion currents at $m/e = 30$ and 28 due to singly ionized $C^{18}O$ and $C^{16}O$ respectively. Here, however, it is necessary to apply a correction, based on the ion current at $m/e = 14$, to the value at $m/e = 28$, because nitrogen from the steel is always present in the gas mixture at the end of a dilution. A more accurate method is to convert part of the carbon monoxide in the equilibrated gas mixture to carbon dioxide, and to measure the ion currents at $m/e = 44$, 46 and 48. The value of R is then given by either $2h_{48}/h_{46}$ or by $(h_{48}/h_{44})^{\frac{1}{2}}$, where h_x represents the ion current at $m/e = x$. The three methods yield results which agree within experimental error. Instrumental errors are avoided by using the same method for both calibration and analysis.

Calibration is effected by admitting to the furnace small measured quantities of normal and enriched oxygen in proportions similar to those encountered in an analysis. Conversion of carbon monoxide to dioxide without isotopic dilution is achieved by compressing part of the gas mixture from the furnace into a small circulatory system which contains a platinum filament heated to 900° C. and a trap cooled in liquid air. By this means the equilibrium :

$$2CO \rightleftharpoons CO_2 + C$$

is constantly displaced until sufficient carbon dioxide has been formed for analysis.

Table 1 summarizes the calibration and analytical results for a standard steel sample, designated *BPL*-21, supplied by the British Iron and Steel Research Association. The mean oxygen content of this steel as determined by several independent investigators with the vacuum-fusion method is given as 0·003 per cent, with an overall range of 0·002–0·004 per cent for individual determinations. Our values are thus higher by a factor of about two. The scatter between individual analyses is not improved.

It is estimated that the combined sources of error in the above analyses should not exceed ± 25 per cent for any single determination. In the analysis of several iron–carbon alloys prepared in this laboratory, results have been duplicated to within 10 per cent in the range 0·006–0·011 per cent oxygen. These figures were again higher than the corresponding vacuum-fusion results by a factor of two to four. The good reproducibility obtained with specimens

Table 1. Isotope Dilution Analysis of Sample *BPL*-21

Sample No.	Weight of sample (gm.)	Volume of enriched oxygen (ml. at N.T.P.)	Volume of normal oxygen used for calibration (ml. at N.T.P.)	$R = {}^{18}O/{}^{16}O$ ($= 2h_{48}/h_{46}$)	Oxygen-18 in enriched gas before dilution (per cent)	Oxygen in sample (per cent)
1	—	0 866	0·634	1·108	90 92	
	5 192	0 716	—	2 820		0 0061
2	—	0·944	0 599	1·200	89·07	
	7·725	0 584	—	1·500		0·0054
3	—	0·658	0 444	1·072	86 52	
	4 178	0 549	—	1·875		0 0063
4	—	0 865	0·589	1·143	89 57	
	4·033	0·478	—	1·311		0 0100
5	—	0·762	0·550	0 932	82 98	
	7·466	0 570	—	0 996		0·0074
					Mean	0 0070

prepared in the laboratory suggests that a large part of the scatter experienced with the British Iron and Steel Research Association standard may be due to inhomogeneities in the sample.

Steels containing manganese may be analysed by this technique if care is taken to introduce the enriched oxygen before the steel sample is dropped into the furnace. This precaution is necessary due to the rapidity with which the ferro-manganese film, which forms on the cold wall of the furnace, reacts with molecular oxygen. Analysis of a British Iron and Steel Research Association standard, designated *BNL*-21, which contained 0·44 per cent manganese, yielded a preliminary value of 0·0196 per cent for the oxygen content. This may be compared with the mean value of 0·0145 per cent reported for this steel by investigators using the vacuum-fusion technique.

Other work in progress in this Laboratory suggests that low results may be obtained for both oxygen and nitrogen by the vacuum-fusion method when the concentration of either of these elements is of the order of 0·01 per cent or less. If this is true, some revision of present accepted thermodynamic data may be necessary for alloys in this range of composition.

A full account of this work will be published elsewhere.

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[1] Kirshenbaum, A. D., and Grosse, A. V., *Trans. Amer. Soc. Metals*, **45**, 758 (1953); *Anal. Chem.*, **26**, 1955 (1954). Kirshenbaum, A. D., Mossman, R. A., and Grosse, A. V., *Trans. Amer. Soc. Metals*, **46**, 525 (1954). Inghram, M. G., "Ann. Rev. Nucl. Sci.", **4**, 81 (1954).

MINERALOGY

Determination of Tetrahedral Aluminium in Mica by Infra-Red Absorption Analysis

Continuation of studies[1] relating the infra-red absorption spectra and the aluminium substitution for silicon in the chlorite minerals has shown that a similar correlation exists in almost all micas. Regardless of the species under study (that is, muscovite, lepidolite, biotite, phlogopite), the shape of the 9–10μ region of the absorption spectra depicts the amount of aluminium substitution for silicon (*Y* number) in the basal, tetrahedrally co-ordinated level in the mica structure. The micas may be natural or synthetic. Position of the maxima of absorption is again dependent upon the dominant type of ion in the octahedrally co-ordinated level, the maxima shifting to slightly longer wave-length with increasing atomic weight of the ions in this position.

The aluminium content and distribution in a mica, therefore, can now be estimated by a rapid physical method. This is in marked contrast to experiences of previous workers[2] who have had great difficulty with differential solubility methods in estimating the aluminium in micas and clays.

In the dioctahedral micas, the aluminium in the octahedral level does not influence the shape of the infra-red absorption pattern, although, because of its lighter mass effect, the position of the maxima is shifted to shorter wave-lengths than with iron- and magnesium-bearing trioctahedral analogues. At first thought, it is strange that such a correlation should exist between *Y* number and absorption spectra, regardless of the mica species present. The tetrahedrally co-ordinated basal level in the mica structure, however, is common to all species; the ions differentiating the species from one another appearing in the octahedral and interlayer positions.

By the courtesy of people too numerous to mention in this short note, who have kindly sent us samples, we have been enabled to include the infra-red absorption curves of 22 analysed micas (Figs. 1 and 2). Members of the same mica species are listed in the vertical columns of curves, in order of decreasing *Y* number, so that horizontal comparison between micas with the same degree of aluminium substitution may be readily made. One of the muscovites and three of the biotites are not yet analysed, but

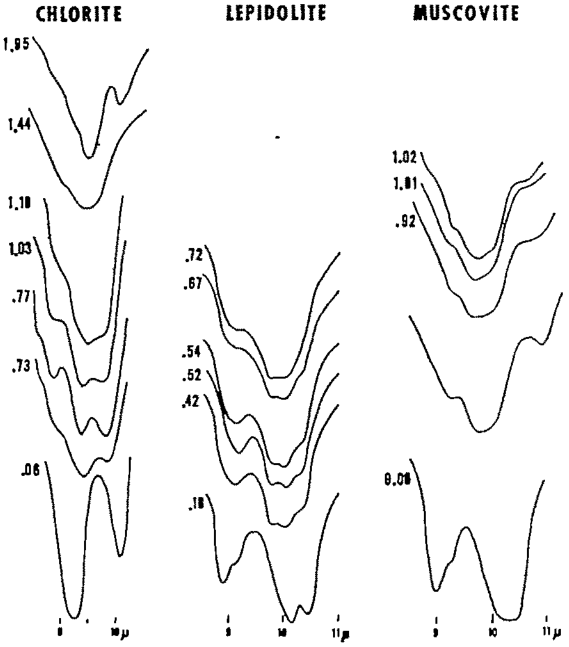

Fig. 1. Centre section (8 5–11μ) of infra-red absorption curves arranged in order of decreasing aluminium substitution for silicon (*Y* number). Curves in each species column are displaced vertically. Micas with equivalent *Y* number may be compared horizontally. Missing number for the muscovite curve is awaiting completion of chemical analysis

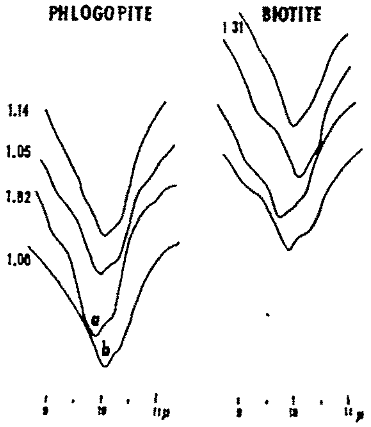

Fig 2. Centre section (8 5–11μ) of infra-red absorption curves for the phlogopite and biotite series. Arrangement as for Fig. 1. Phlogopite curves (*a*) and (*b*) have same Y number but are displaced laterally 0 2μ. Curve (*a*) is a magnesian phlogopite whereas (*b*) has a 1/8 iron substitution. Three of the biotites are not analysed at the present time

have been placed in relative position as deduced from the shape of their curves.

As with the chlorite series, the pure tetrasilicic end member ($Y=0$) (of at least the muscovite and lepidolite series) has a complex absorption pattern in the 9–10μ region. In addition to the normal broad absorption (or doublet) at 10·1–10·4μ, a pronounced doublet appears between 8·85 and 9 04μ. With increasing aluminium substitution, as Y rises from 0·4 towards 0·6, this doublet between 9·8 and 10·3μ becomes a triplet, with the short wave-length peak of this triplet becoming dominant as Y values increase above 0·9. The doublet at 9μ gradually fades out as Y increases towards unity, leaving the simplified, single absorption at 9·7–9·8μ.

Only one mica sample with Y value greater than 1·2 could be obtained. It may be that this value of 1·2 is again as critical in the broad groups of micas as it was in the chlorites[1], marking some type of structural change.

Difficulty has been experienced in relating the minerals talc and pyrophyllite to this scheme. Both possess effectively a zero aluminium substitution ($Y=0$), but their absorption spectra, which are very similar in pattern, consist of simple, single absorptions, suggesting a $Y=2$ situation as in the chlorite series. In addition, margarite, with a $Y=2$, has an unusually complex pattern and is markedly displaced to higher wave-lengths, compared with the septeamesite ($Y=2$) of the chlorite study.

It would be expected that the montmorillonite groups would show a similar correlation between Y number and infra-red absorption, although their lower degree of crystallinity would make the individual absorptions less distinct. Investigation of all these points is actively progressing.

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[1] Tuddenham, W. M., and Lyon, R. J. P., *Anal Chem*, 31, 377 (1959)

[2] Yoder, H. S., in "Clays and Clay Minerals", Monograph No. 2 (Pergamon Press, New York, 1959).

CHEMISTRY

The α- and β-Forms of Calcium Sulphate Hemihydrate

Kelley, Southard and Anderson[1] have shown that the thermodynamic properties of calcium sulphate hemihydrate depend on whether the hemihydrate is produced from gypsum calcined in a steam-saturated atmosphere (α-hemihydrate), or under conditions where the partial pressure of water vapour is low (β-hemihydrate), but the nature of a hemihydrate prepared under conditions intermediate between these two extremes (for example, the manufacture of gypsum plaster by the kettle process) is not known for certain. These authors considered that such a hemihydrate would consist of a mixture of the α- and β-forms, but Eipeltauer[2] considered that there would also be some hemihydrate crystals having properties intermediate between these two forms. Work that I have done shows that only the α- and β-hemihydrates can be detected by differential thermal analysis in a sample of hemihydrate prepared by calcining gypsum under conditions simulating the manufacture of gypsum plaster in a kettle.

By means of differential thermal analysis it is possible to detect in a sample of calcium sulphate hemihydrate the various forms the rates of transformation from soluble to insoluble anhydrite of which vary[3], since each phase-change is shown by an exothermic peak at a particular temperature on the differential thermal curve. A sample of hemihydrate was prepared by heating 800 gm. of a washed natural gypsum for 5 hr. in a glass container filled to a depth of 3 in. (oven temp. 170° C.). The sample was cooled and then dried at 50° C., and the water content determined as 6·05 per cent (theoretical 6·21 per cent). The differential thermal curve for a partly dehydrated sample of this hemihydrate is shown in Fig. 1, curve *a*. The water content of the sample was reduced to about 1 per cent by heating for 1 hr. at 120° C., otherwise the exotherm shown at about 245° C. was completely absorbed in the large endotherm due to the loss of water from hemihydrate. After heating at 120° C. duplicate samples were allowed to rehydrate at 65 per cent relative humidity; resultant changes in weight indicated that the amount of insoluble anhydrite formed at 120° C. was very small. The exotherm shown at 245° C. on curve *a*

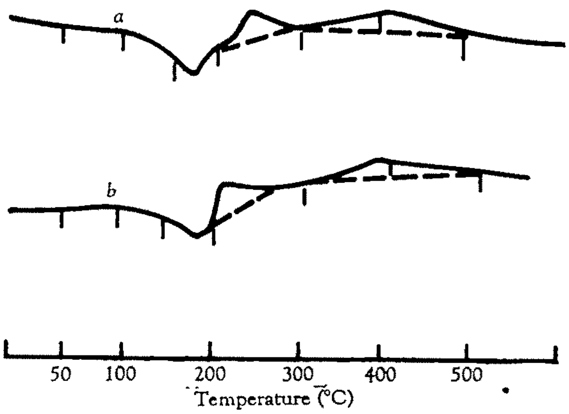

Fig 1. Differential thermal analysis curves for calcium sulphate hemihydrate (water content 1 per cent). *a*, a hemihydrate prepared under conditions simulating manufacture of gypsum plaster by the kettle process. *b*, a mixture of 70 per cent α-hemihydrate and 30 per cent β-hemihydrate

appears to be complete before the second exothermic reaction commences at about 300° C., and there is no indication of the decomposition of a phase intermediate between these two. The size and shape of the two exothermic peaks on curve *a* are very similar to those shown on curve *b*, which was obtained from a mixture of 70 per cent of α-hemihydrate, prepared by autoclaving, and 30 per cent β-hemihydrate. For comparison, the water content of this sample was also reduced to about 1 per cent prior to differential thermal analysis. The peak temperatures of the two exotherms on curve *a* occur at slightly higher temperatures than those on curve *b*, and the exotherm at 210° C. on the latter curve appears to commence before the small endotherm due to the loss of about 1 per cent water is complete.

The differential thermal analysis of a hemihydrate prepared as described above shows the presence of two main phases only, and by this technique it was not possible to detect a phase intermediate between the two. However, since the peak temperatures of the two exotherms on curve *a* occur at slightly higher temperatures than those for the mechanical mixture (curve *b*), this may indicate slight differences between different preparations of both α- and β-hemihydrates.

In general it was found that the proportion of each phase shown on the differential thermal curve could be determined more accurately from the area beneath the β-exotherm, because any insoluble anhydrite that might form during the preparation of a sample would be produced mainly from the more reactive α-phase. Since it has been shown that the α-exotherm is due only to the first rapid part of the transformation of α-soluble anhydrite to insoluble anhydrite[3] (the latter part of the reaction is slow and is not recorded by differential thermal analysis) the formation of small amounts of insoluble anhydrite prior to differential thermal analysis can greatly reduce the size of the α-exotherm.

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[1] Kelley, K. K., Southard, J. C., and Anderson, C. T., U.S. Bur. Mines Tech. Paper 625 (1941).

[2] Eipeltauer, E., *Zement-Kalk-Gips*, 9, 501 (1956).

[3] Powell, D. A., *Nature*, 182, 792 (1958).

Molybdenum and Niobium Sulphides

In the system Mo-S the only compound that is formed by direct synthesis from the elements is the disulphide MoS_2. This compound exists in two modifications:

(1) The well-known hexagonal form ($a=3·16$; $c=12·29$ A.), the structure of which[1] is shown in Fig. 1*a*.

(2) The rhombohedral form ($a=3·17$; $c=18·38$ A.) which was first described by Bell and Herfert[2]. These authors propose a $CdCl_2$-type structure which, however, is incompatible with their own and our X-ray diffraction data. Good agreement is found for the structure shown in Fig. 1*b*. This structure can be described in space-group $R3m—C^5_{3v}$: 3 Mo situated in 3(*a*) : 0 0 *z*, etc., with $z=0$; 3 S I in 3(*a*) with $z\approx\frac{1}{4}$ and 3 S II in 3(*a*) with $z\approx 5/12$.

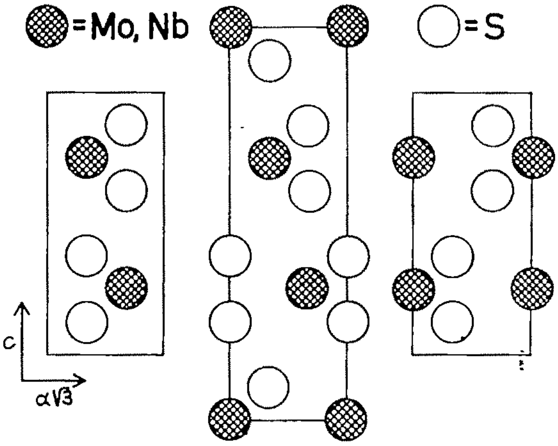

Fig. 1. Sections [through the (11$\bar{2}$0) planes of (*a*) hexagonal MoS_2, (*b*) rhombohedral MoS_2 and NbS_2, (*c*) hexagonal NbS_2

In both forms of MoS_2 the metal environment is trigonal prismatic. In synthetic molybdenum disulphides generally a considerable degree of stacking disorder in the *c* direction is present which causes a weakening of the reflexions with $h-k\neq 3n$, increasing with increasing value of *l*.

In the system Nb-S the following phases are observed:

(1) Monoclinic NbS_3 ($a=4·94$; $b=6·74$; $c=18·1$ A.; $\beta=97·5°$), which is probably isotypic with TiS_3 [3] and $ZrSe_3$ [4].

(2) Rhombohedral NbS_2 ($a=3·33$; $c=17·81$ A.), stable below about 800° C. This phase is probably identical with the disulphide found by Hägg and Schönberg[5], for which they propose a $CdCl_2$-type structure. This structure type, however, is in conflict with our intensity data; the structure we derive is isotypic with rhombohedral MoS_2 (Fig. 1*b*). Generally, stacking disorder in the *c* direction is observed; it can be diminished by annealing at 700–800° C.

(3) Hexagonal NbS_2 ($a=3·31$; $c=11·89$A.) is formed from the elements above 850° C. and is stable up to 1,050° C. The space group is $P6_3/mmc—D^4_{6h}$, the same as in the case of hexagonal MoS_2, but the structure is different. 4 S lie in 4(*f*) : $\pm(\frac{1}{3}, \frac{2}{3}, z; \frac{1}{3}, \frac{2}{3}, \frac{1}{2}-z)$ with $z\approx\frac{1}{8}$; 2 Nb either in trigonal prismatic holes 2(*b*) : $\pm(0\ 0\ \frac{1}{4})$ or in octahedral holes 2(*a*) : $0\ 0\ 0$; $0\ 0\ \frac{1}{2}$. X-ray diffraction can scarcely distinguish between these two arrangements, which are exactly homometric if $z_S=\frac{1}{8}$. Because of the analogy with rhombohedral NbS_2 and because of stacking arguments, position 2(*b*) is preferred (Fig. 1*c*). Once more, stacking disorder in the *c* direction is found.

(4) Rhombohedral $Nb_{1+x}S_2$ ($a=3·33$; $c=17·80$–$17·85$ A.), the range of homogeneity of which stretches from $x\approx 0·12$ to $x\approx 0·25$ at 1,100° C. and to $x\approx 0·5$ at 800° C. The structure which is shown in Fig. 2*a* can be described in space group $R3m—C^5_{3v}$: 3 Nb I in 3(*a*) : 0 0 *z*, etc., with $z=0$; $3x$ Nb II in 3(*a*) with $z\approx 5/6$; 3 S I in 3(*a*) with $z\approx\frac{1}{4}$; 3 S II in 3(*a*) with $z\approx 5/12$. The co-ordination of Nb I is trigonal prismatic, that of Nb II octahedral. Here, stacking disorder is inhibited by the presence of Nb II between the layers.

(5) Hexagonal $Nb_{1+x}S_2$ ($a=3·31$; $c=12·53$–$12·65$ A.) is stable above about 1,000° C. in the range $x\approx 0·30$ to $x\approx 0·43$. The space group is $P6_3/mmc—D^4_{6h}$; 2 Nb I are situated in 2(*b*) : $\pm(0\ 0\ \frac{1}{4})$; $2x$

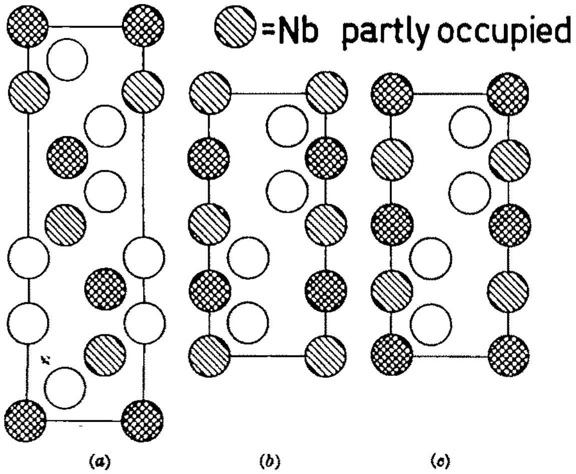

Fig. 2. Sections through the (11$\bar{2}$0) planes of (a) rhombohedral $Nb_{1+x}S_2$, (b) hexagonal $Nb_{1+x}S_2$, (c) one of the possible arrangements of $Nb_{2-y}S_2$

Nb II in 2(a) : 0 0 0 ; 0 0 $\frac{1}{2}$ and 4 S in 4(f) : $\pm(\frac{1}{3}, \frac{2}{3}, z;\ \frac{1}{3}, \frac{2}{3}, \frac{1}{2}-z)$ with $z \approx \frac{1}{8}$. This structure (Fig. 2b) seems more likely than the homometric arrangement with interchanged Nb I and Nb II positions, the reasoning being the same as in the case of hexagonal NbS_2. Again, stacking disorder is prevented by the presence of Nb II.

(6) Hexagonal $Nb_{2-y}S_2$ is stable below about 850° C. in the range $y \approx 0{\cdot}0$ to $y \approx 0{\cdot}3$. The X-ray patterns closely resemble those of hexagonal $Nb_{1+x}S_2$ and, therefore, the structure is either analogous to that of hexagonal $Nb_{1+x}S_2$ (Fig. 2b) with statistical vacancies in the 2(a) site, or to its homometric form with the vacancies in the 2(b) position (Fig. 2c). The possibility that hexagonal $Nb_{1+x}S_2$ and $Nb_{2-y}S_2$ belong to the same homogeneity range cannot be excluded. $Nb_{2-y}S_2$ is probably identical with the phase NbS described by Hagg and Schonberg[5]. These authors propose an NiAs-type structure for NbS with an excess of sulphur ($a=3{\cdot}32$; $c=6{\cdot}46$ A.) and a WC-type structure for NbS in equilibrium with Nb ($a=3{\cdot}32$; $c=3{\cdot}23$ A.). Evidently, Hägg and Schönberg overlooked the reflexions with $l \neq 2n$ which have rather weak intensities ; for $y=0$ the intensities of the reflexions with $l=4n+2$ become zero. Therefore, the c axes found by Hägg and Schönberg[5] are one-half and one-quarter of the actual one respectively.

(7) Above 850° C. one or more phases occur at compositions about NbS. The relationships in this range are rather complicated and not yet fully understood.

Details of these investigations will be reported elsewhere.

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[1] Dickinson, R G , and Pauling, L., *J. Amer. Chem. Soc.*, 45, 1465 (1923)

[2] Bell, R. E., and Herfert, R. E., *J. Amer. Chem. Soc* , 79, 3351 (1957).

[3] Hahn. H., and Harder, B. *Z. anorg. Chem.*, 288, 241 (1956).

[4] Kronert, W., and Plieth, K., *Naturwiss.*, 45, 416 (1958).

[5] Hägg, G., and Schönberg, N., *Arkiv Kemi*, 7, 371 (1954)

Oscillographic Investigation of Electrodeposition of Chromium

A great deal of work on the theory of electrodeposition of chromium has been carried out since 1920. E. Müller[1] reported the formation of chromium chromate film on the cathode surface and inferred the mechanism of chromium electrodeposition from the polarization phenomena. Recently D. Reinkowski and C. A. Knorr[2] fully examined the reproducibility of the cathode-polarization curves, and presented a new view of the existence of their different potential range and on the transition between these ranges. More recently, H. Gerischer and N. Käppel[3] postulated another theory of the chromic oxide film formed on the cathode surface, measuring the quasi-stationary potential-current curves using a potentiostatic method.

In the present communication, the charging curves during cathodic polarization at various current densities in the solution containing mainly 3 *M* CrO_3 were measured by the oscillographic method devised by A. Hickling[4]. The electrolytes used in this experiment were as follows: (a) 3*M* CrO_3, (b) 3 *M* CrO_3, 0·03 *M* sulphuric acid. The electrolytic temperature was always kept at 20° C. The charging curves at the platinum and gold electrodes during cathodic polarization at various current densities in the solution (b) are shown in Figs. 1*A* and 2*A*, respectively. The schematic representations of these curves at 20 amp./sq. dm. are shown in Figs. 1*B* and 2*B* respectively. The ordinates represent the potentials vs. *S.C.E.* and the abscissæ are proportional to the quantities of electricity passed. In solution (a), at all current densities the charging curves were the same tracks as those obtained in the solution (b) at 20 amp./sq. dm.

The tracks of the charging curves at 20 amp./sq. dm. in solution (b) passed through four inflexion points (1, 2, 3 and 4 from positive potentials to negative), and reached the constant potentials, forming the three stages (a, b and c), as shown in Figs. 1*B* and 2*B*.

In comparison with the current-potential curves in solution (b) at both electrodes (see Figs. 5 and 7 of ref. 2), which consisted of four or three different potential-ranges, it may be noted that the inflexion points (2) (+192 mV. at the platinum electrode, +674 mV. at the gold electrode) approximately correspond to the ends of the first stages of the polarization curves, in the same manner (3) (−268 mV. (platinum) +296 mV. (gold)) to the ends of the second stages, (4) (−412 mV. (platinum), −780 mV. (gold)) to the beginning of the third or fourth stages. At the lower current densities (5 amp./sq. dm.,

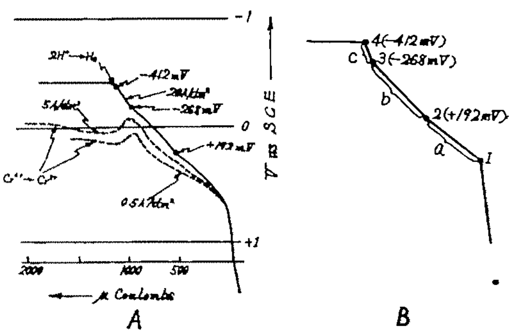

Fig. 1. *A*, Charging curves of the platinum electrode during cathodic polarization in the solution (b) at various current densities ; *B*, schematic representation of Fig. 1 at 20 amp./sq. dm.

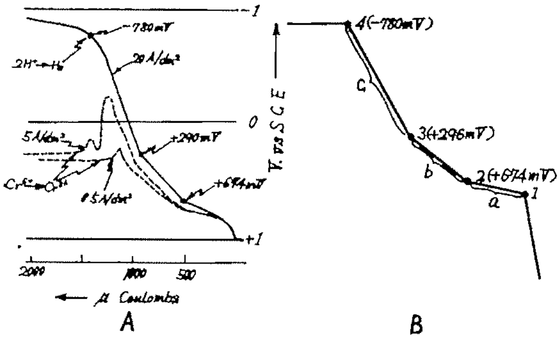

Fig. 2, *A*, The charging curves of the gold electrode during cathodic polarization in the solution (*b*) at various current densities, *B*, the schematic representation of Fig. 2 at 20 amp./sq. dm.

0·5 amp./sq. dm.) in solution (*b*) the tracks of the oscillograms did not reach the inflexion points (4), but gave constant potentials at the inflexion points (3) or (2). At these potentials only the reduction process of Cr^{6+} to Cr^{3+} occurred.

A. Hickling[4,5] and S. E. S. El Wakkad *et al.*[6,7] studied the reduction of the oxide films of platinum and gold electrodes, measuring the charging curves of these electrodes during cathodic polarization. In comparison with their results (see Fig. 9 of ref. 6 and Plate II, I of ref. 5), it is seen that the stages (*a*) in Figs. 1 and 2 correspond to those of the reduction processes of PtO and Au_2O_3, respectively. F. Baumann and I. Shain[8] found that the reduction process of Cr^{6+} to Cr^{3+} occurred soon after the reduction of the oxide film of the gold electrode, using a gold rotating electrode. Consequently, it is considered that at stage (*a*) the reduction of the oxide films of these electrodes occurs.

From the fact that the stage (*b*) exhibited the same slope at both electrodes, that is, platinum and gold, it is suggested that the same electrode reaction occurs. The inflexion points (3) corresponded to the ends of the second stages on the polarization curves, and at the ends of these stages the electrodes should be fully covered with chromium chromate film. Therefore, it is considered that stage (*b*) exhibits the quantity of electricity required for the formation of this film. The number of ions of Cr^{3+} formed on the stage (*b*) were evaluated from the quantity of the electricity: at the platinum and gold electrodes these are $1{\cdot}00 \times 10^{15}$ ions/sq. cm. and $1{\cdot}37 \times 10^{15}$ ions/sq. cm., respectively. The number of atoms of platinum and gold per sq. cm. at the platinum and gold surfaces are, respectively, $1{\cdot}61 \times 10^{15}$ atoms/sq. cm. and $1{\cdot}50 \times 10^{15}$ atoms/sq. cm. Considering the uncertainty of the exact values of the surface areas of both electrodes, it may be considered that the quantities of electricity necessary for the formation of the films approximately correspond to the formation of a unimolecular film of chromium (III) on the electrodes, and this quantity of electricity at the platinum electrode is in fair agreement with the value obtained by I. M. Kolthoff and A. M. S. El Din[9].

The significance of the four stages of the polarization curve in the solution (*b*) (chromium plating solution) has not been fully explained; and D. Reinkowski and C. A. Knorr[2] have indicated that the second stage exists in the different potential range depending on the electrode material.

From the above results, it is considered that at the first stage of the polarization curve the reduction of the oxide film of the electrode occurs in the case of platinum and gold electrodes, and that the film of chromium chromate forms soon after this reduction process occurs. Consequently, the fact that the first and the second stages of the polarization curve exist at the different potential ranges, depending on the electrode materials, was attributed to the differences in the reduction potentials of the oxide films of the electrodes.

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[1] Muller, E., *Z. Elektrochem.*, 32, 399 (1926); 50, 172 (1944).
[2] Reinkowski, D., and Knorr, C. A., *ibid.*, 58, 709 (1954).
[3] Gerischer, H., and Käppel, M., *ibid.*, 61, 463 (1957).
[4] Hickling, A., *Trans. Faraday Soc.*, 41, 333 (1945).
[5] Hickling, A., *ibid.*, 42, 518 (1946).
[6] El Wakkad, S. E. S., and Emara, S. H., *J. Chem. Soc.*, 461 (1952).
[7] El Wakkad, S. E. S., and Emara, S. H., *ibid.*, 3098 (1954).
[8] Baumann, F., and Shain, I., *Anal. Chem.*, 29, 303 (1957).
[9] Kolthoff, I. M., and El Din, A. M. S., *J. Phys. Chem.*, 60, 1564 (1956).

A Synergic Method of carrying out Continuous Electrochromatography

ALTHOUGH some workers using the technique of continuous electrochromatography on conventional anticonvection media (such as filter paper or glass powder) have supposed that specific adsorption effects play a part in the separation, the extent to which these can in fact influence the separation, if at all, is negligible. The adsorbed Stern layer ions are practically immobile with respect to both electrophoretic and hydrodynamic gradients; thus even notionally, since each is equally affected, the resultant of motion in the two mutually perpendicular directions should be independent of the degree of adsorption. Weber[1] has considered the general differential equation describing the concentration distribution (as a function of space and time) in terms of diffusion, adsorption, and the two velocities, on the assumption that adsorption equilibrium is reached instantaneously. The solution of this equation for the stationary state does not involve an adsorption term.

It seems likely that improved electrochromatographic separations might be achieved by substituting ion-exchange membrane 'curtains' for paper ones. In this way a stationary phase capable of highly selective sorption could be provided, within which particles would be immobilized with respect to eluant flow but not with respect to current flow. Consequently the change brought about in mobility ratios might be expected to differ from that brought about in retention-volume ratios; and in certain systems at least the direction of these changes might be such as to give rise to synergy between electrophoresis and chromatography. An analysis of the situation shows that this expectation is well founded. If we define a 'separation coefficient' $D^1{}_2$ as the ratio $\tan\theta_1/\tan\theta_2$, where θ is the angle through which the path of a given species is swept from the vertical under the influence of an applied potential, then the following relations can be derived:

Ion-exchange resin absent:

$$D^1{}_2 = \frac{u_1}{u_2} \cdot \frac{\rho^0{}_2}{\rho^0{}_1} \qquad (1)$$

Ion-exchange resin present (volume of eluant per gm. resin small):

$$D^1{}_2 = \underbrace{\frac{\bar{u}_1}{\bar{u}_2}}_{\text{electrophoretic factor}} \cdot \underbrace{\frac{\rho^r{}_2}{\rho^r{}_1} \cdot \frac{K^0{}_1}{K^0{}_2}}_{\text{chromatographic factor}} \qquad (2)$$

The first term in each case is a ratio of mobilities, measured in the free eluant in (1) and in the resin in (2). The third term in (2) is the ratio of specific retention volumes, measured in the *absence* of any complexing agent in the eluant. The second term arises from the possible presence of a ligand. The quantity ρ_i is the ratio (total concentration of species i in eluant)/(concentration of uncomplexed species i in eluant), and depends on the concentrations of both i and ligand as well as pH. The superscripts denote presence or absence of ion-exchange resin, since this factor is altered by the equilibria displacements resulting from the introduction of resin. In the derivation it is assumed that the mobility of the complexed species is negligibly small in comparison with that of the uncomplexed species, that diffusion can be neglected and exchange equilibria regarded as instantaneous, and that the resin is lightly loaded.

Comparison of equations (1) and (2) shows at a glance why synergy arises when an ion-exchanger is introduced into the system. In general one can say, for a series of like ions, that the greater the mobility of an ion the greater its affinity for an ion-exchange resin and the lower the stability of its complexes[2]. Furthermore it can be shown that the 'complexone affinity' term in equation (2) is very much further from unity than that in equation (1) for a given total concentration of complexone, unless vanishingly small quantities of material are to be separated. (In the absence of complexone it is, of course, unity.) This is an important effect *per se*.

A suitable arrangement for separations of this kind consists of a pack of ion-exchange membranes interleaved with filter paper or similar material to act as flow stabilizer. This has the advantage over a trough filled with fine resin granules in that it is relatively easy to obtain uniform flow. Moreover, membranes of high selectivity are likely to become more readily available than their granular counterparts. For a given system the coefficient $D^1{}_2$ can be evaluated most simply by carrying out separate electrophoretic and chromatographic separations on strips of paper or paper–membrane–paper sandwich. Such strips are readily examined spectrophotometrically or radiometrically, and it is only necessary to divide the ratio x_1/x_2 of distances moved in an electrophoretic run by the corresponding ratio for a chromatographic run carried out under the same conditions. Runs have been performed on single strips of sulphonated polyvinyl chloride membrane[3] 3 cm. wide and 30–45 cm. long sandwiched between two strips of 'Separa *DHC*' paper (W. and R. Balston, Ltd.) of the same width. For convenience the system sodium-22 (2·6 yr.)–potassium-42 (12·4 hr.) was chosen for study. The membranes were used in the dimethylammonium form; the eluant (electrolyte) contained uramil-N,N-diacetic acid and dimethylamine, its pH being determined by the concentration of the latter. Similar conditions have been employed successfully to resolve mixtures of alkali ions on ordinary ion-exchange columns[4]. The electrophoretic runs required about 0·04 ampere-hours at 750 volts.

The results obtained in a series of experiments with the membrane are compared with those for paper alone in Fig. 1. The strips were loaded with 0·5 μmole of each ion. It is seen that although the mobility ratio is somewhat decreased in the presence of membrane its pH dependence parallels the behaviour in the absence of membrane. On the other hand the corresponding chromatographic ratio, in the presence of membrane, undergoes a sharp decrease between pH 8 and 9. This can be traced to the operation of the complexone affinity term in equation (2). Although in this system the membrane itself exhibits no noticeable préference, the synergic influence of the chromatographic component in its presence is evident.

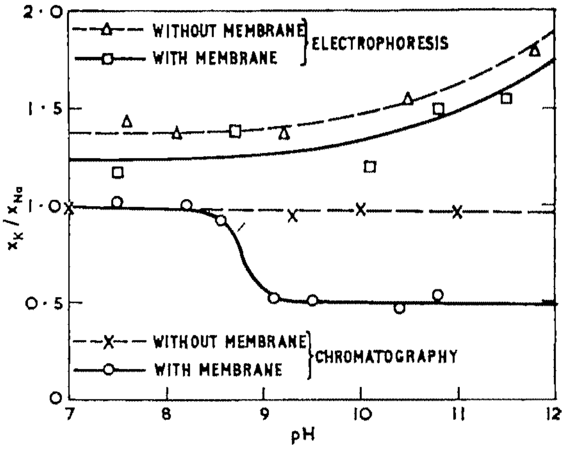

Fig. 1. Evaluation of a system by means of strip experiments

A full account of this work, including a comparison of several membranes and a description of a working cell used for the separation of lithium, sodium and potassium, is in preparation.

Acknowledgement is gratefully made to Mr. T. A. Gough for technical assistance. This note forms the substance of Brit. Pat. Appl. No. 39033/58, and is published by permission of the National Research Development Corporation and the Director, National Chemical Laboratory.

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[1] Weber, R., *Helv. Chim. Acta*, 36, 424 (1953).

[2] Reichenberg, D., and McCauley, D. J., *J. Chem. Soc.*, 2741 (1955). Reichenberg, D., *Chem. Technik*, 10, 454 (1958).

[3] Hookway, H. T., Hale, D. K., and Goldsmith, R. G., Brit. Pat. Appl. 830 (1957).

[4] Buser, W., *Helv. Chim. Acta*, 34, 1635 (1951).

Reduction of Graphite by Lithium-Ethylenediamine

It is well known that coals can be converted to liquid fuels by catalytic hydrogenation at high temperatures and pressures. However, coals of high rank (>89 per cent carbon m.a.f.) are liquefied in poor yield[1]; anthracites (>92 per cent carbon m.a.f.) are very difficult to hydrogenate; and graphite cannot be hydrogenated to liquid or solid products.

Recent work in this laboratory[2] has shown that coals and vitrains containing up to 92 per cent carbon can be reduced by the lithium-ethylenediamine system[3]; hydrogen is added, no carbon–carbon bonds are broken, and the reaction takes place under the mild conditions of 115° and atmospheric pressure.

Table 1

	C	H	N	Ash	O (diff.)	gm. H_2 added per 100 gm.	gm. H_2O added per 100 gm.
Graphite, starting material*	98·90	0·29	0·07	0·87	−0·13		
Graphite, reduced*	85·39	2·91	3·83	6·28	1·59		
Graphite, starting material†	99·79	0·29	0·07	—	−0·15		
Graphite, reduced†	95·95	2·12	0·07	—	1·86	1·65	2·85

* Actual analytical values.

† Analytical values corrected to a moisture-, ash- and ethylenediamine-free basis

The insolubility of graphite in ethylenediamine should hinder its reduction; on the other hand, it is already known that even an insoluble anthracite is reduced by lithium-ethylenediamine; further, the graphite structure consists almost entirely of carbon–carbon single and double bonds, and the latter are readily reduced by lithium-ethylenediamine.

Three gm. of Madagascar flaky graphite (previously de-ashed by treatment with hydrofluoric acid) was reduced with 6·80 gm. of lithium and 150 ml. of ethylenediamine[2]. The recovered material weighed 3·11 gm. Table 1 gives the analytical values for the product. The graphite after reduction retains nitrogen, probably in the form of chemically combined ethylenediamine; this is the same behaviour found with coal. A correction must be made for this combined ethylenediamine, since it contains hydrogen. Table 1 gives the analytical composition of the graphite, corrected to a moisture-, ash-, and ethylenediamine-free basis. The value of 1·65 gm. of hydrogen added per 100 gm. of starting graphite seems remarkably high, particularly when the mildness of the conditions used is considered. This value of 1·65 gm. corresponds to addition of 19·7 atoms of hydrogen per 100 carbon atoms in the original graphite. This is much lower than the value of 44·8 atoms of hydrogen per 100 carbon atoms found for a high-rank bituminous vitrain[3] (90·4 per cent carbon m.a.f.), but it compares favourably with the value of 16·9 found for an anthracite[3].

To determine the effect of the treatment on the graphite lattice, X-ray diffraction patterns of the treated sample were examined. The patterns of the untreated and treated graphite (Fig. 1) were obtained from compressed slab specimens by the reflexion method. The (002) band of the treated sample was greatly decreased (not obvious on Fig. 1) and broadened and has a peculiar shape. Furthermore, a distinct band has been formed at a *d*-spacing of 4·82 A. Lithium is known to form interstitial compounds with graphite. The reduced sample was analysed and found to contain 1·5 per cent lithium. To decrease the lithium content, the sample was treated with hydrochloric acid and washed. It then contained 0·47 per cent lithium. The X-ray patterns of the sample before and after this treatment were identical; that is, the (002) and 4·82 A. bands were not affected. The large amount of nitrogen present is somewhat disturbing; however, it appears that hydrogen atoms have been added to the graphite lattice to produce hydrogenated layers. The average distance between parallel adjacent alicyclic units is estimated to be 4·7–4·9 A.

X-ray examination of the compressed slab specimen by the transmission method revealed that the (*hkl*) reflexions were also reduced and broadened. This suggests that treatment of graphite flakes with lithium-ethylenediamine effectively reduces the size of crystals, in particular the stacking height.

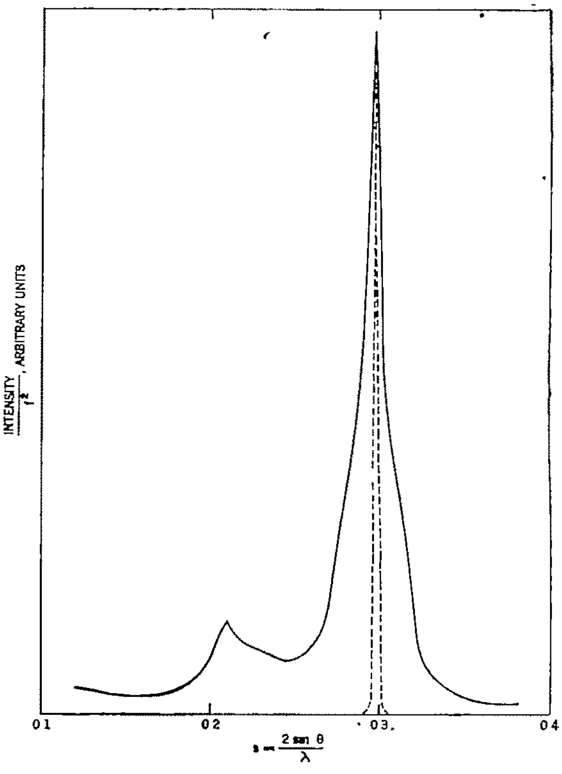

Fig. 1

We wish to thank V. Tiensuu for assistance in the interpretation of the X-ray patterns.

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[1] Fisher, C. H., Sprunk, G. C., Eisner, A., O'Donnell, H. J., Clarke, L., and Storch, H. H., Bureau of Mines Technical Paper 642, 44 (1942), and references cited therein.

[2] Reggel, L., Raymond, R., Friedman, S., Friedel, R. A., and Wender, I., *Fuel*, 37, 126 (1958). Reggel, L., Raymond, R., and Wender, I., Abstracts of Papers, Division of Gas and Fuel Chemistry, 135th Meeting, American Chemical Society, Boston, Mass. (April 1959).

[3] Reggel, L., Friedel, R. A., and Wender, I., *J. Org. Chem.*, 22, 891 (1957). Reggel, L., Friedman, S., and Wender, I., *ibid.*, 23, 1136 (1958).

BIOCHEMISTRY

Violinoid Products of Oxidation of *d*-Urobilin

Some confusion has recently arisen over the nature of the pigments derived from *d*-urobilin (I) by treatment with methanolic ferric chloride in the presence of hydrochloric acid. Watson[1] claims that this urobilin, after treatment for 15 min. under conditions similar to those described below, affords mainly glaucobilin (II). This finding differs from our own[2], which suggested that mesobiliviolins were the main products. Watson obtained mesobiliviolin only after brief treatment of *d*-urobilin with ferric chloride.

We have now confirmed that our original conditions of oxidation[2] convert *d*-urobilin mainly to violinoid pigments (λmax. 560–600 mμ in 2·8 *N* hydrochloric acid, 570 mμ in chloroform and 630,580 mμ for the zinc complex in methanol; these bands being

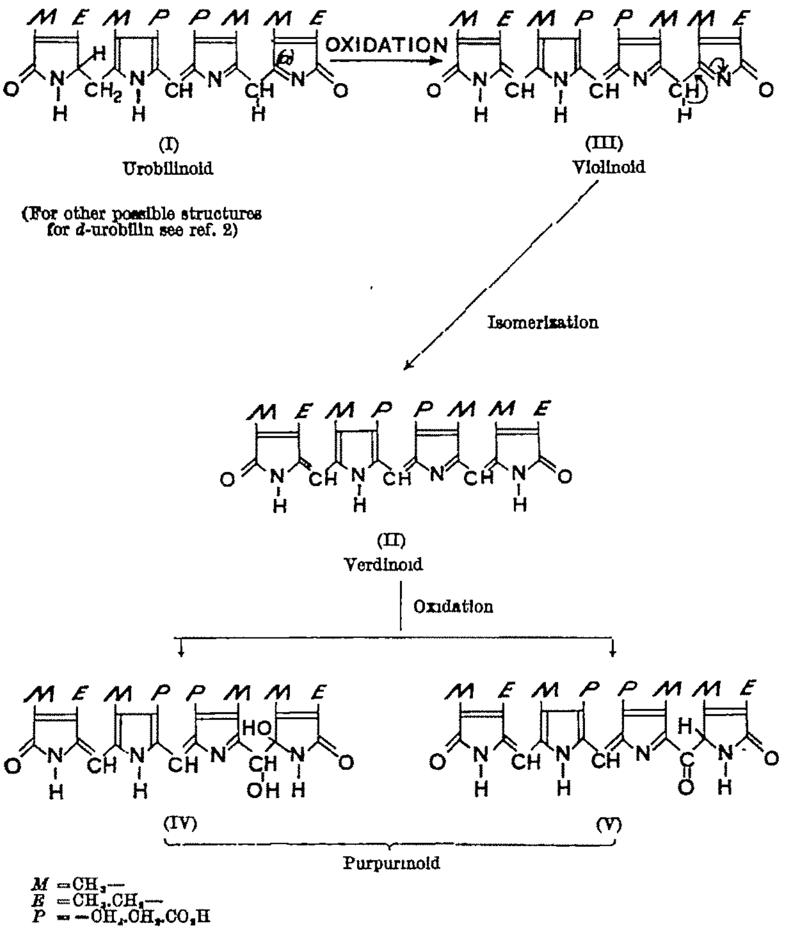

Fig. 1

diminished with formation of a third band at 510 mμ on addition of iodine). We have also found that further treatment with ferric chloride converts these violins to glaucobilin, so that they must be true violins, that is, bilidienes (III)[3]; and not the spectroscopically similar bilipurpurins[4], that is, compounds like (IV) and (V) derived from verdins (II) by oxidative saturation of one methene group. Further study of the problem has shown that even minor changes in our procedure can, according to conditions, give glaucobilin, mesobiliviolin, mesobilipurpurin or a mixture of these pigments. The nature of the products depends, among other things, upon the time of ferric chloride oxidation, dilution of the reaction mixture and extraction of products.

200-μgm. quantities of *d*-urobilin hydrochloride, with $(\alpha)_D$ (in chloroform) +5,000, were obtained by evaporation of appropriate volumes of chloroform solution. After solution in 2 ml. methanol the pigment was boiled, under reflux, with 1 ml. of 25 per cent (w/v) solution of ferric chloride in 11 *N* hydrochloric acid. After addition of 4 ml. of saturated aqueous sodium acetate solution and sometimes dilution, the oxidation products were extracted into ether. The pigments were taken from ether into 2·8 *N* hydrochloric acid; absorption spectra of the acid solutions were measured and the pigments were extracted into chloroform. After washing and filtration the spectra were again determined.

When the solutions were boiled for 10–15 min. and the pigments extracted without dilution, glaucobilin accompanied by small quantities of violin was formed, as was apparent from the absorption spectra. On refluxing for 15–18 min. the reaction mixture, still initially green, on dilution rapidly (1–5 min.) acquired a pink-violet colour due to the formation of violinoid pigments (λmax.: 540–560 mμ in chloroform, 580–590 mμ in 2·8 *N* hydrochloric acid and 620, 575–585 mμ for the zinc complex in methanol; on addition of iodine to the last solution there was depression of the existing two peaks, and an additional peak appeared at 510 mμ). These pigments would not undergo further oxidation to verdin on treatment with ferric chloride and were therefore assumed to be purpurins rather than true violins; the behaviour of their chloroform solution on treatment with 1 per cent hydrochloric acid suggests that they are in fact mesobilipurpurins-II (for example, V)[4]. Refluxing for 30 min. gave a reaction mixture from which glaucobilin alone could be isolated independently of dilution or interval between end of reaction and extraction of product.

It is evident that glaucobilin formation by oxidation with ferric chloride is more rapid than the corresponding reaction with *i*-urobilin. We suggest that a possible reason for this is that violin, initially formed by dehydrogenation of one bridge carbon atom and its adjacent α-pyrrole carbon atom, undergoes rapid isomerization to verdin by the prototropic change formulated in Fig. 1. A similar change is impossible for *i*-urobilin since it lacks the necessary unsaturation (*a*) present in *d*-urobilin[2].

In agreement with this mechanism is the fact that *d*-urobilin can be isomerized to a true violin, unaccompanied by glaucobilin, by anaerobic treatment with alkoxides; these are effective catalysts for changes such as those shown in Fig. 1[5,2]. The mechanism is, however, not in accord with a recent observation of Watson (private communication) that in ferric chloride oxidation *d*-urobilin IXα behaves more like *d*-urobilin than *i*-urobilin. If this observation is confirmed, the structures postulated for *d*-urobilin and *d*-urobilin-IXα[2] may need revision, for the latter has hitherto been considered optically isomeric with *i*-urobilin.

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[1] Watson, C. J., and Weimer, M., *Fed. Proc.*, 18, No. 1377 (1959); and Watson, C. J., private communication.

[2] Gray, C. H., and Nicholson, D. C., *J. Chem. Soc.*, 637, 3085 (1958).

[3] Siedel, W., *Z. physiol. Chem.*, 237, 8 (1935).

[4] Lemberg, R., and Legge, J. W., "Hæmatin Compounds and the Bile Pigments", 130–133 (Intersci. Pub., New York, 1949).

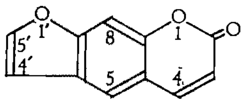
[5] Baker, J. W., "Tautomerism", 51 (Routledge, London, 1939).

Activating and Fluorescent Wave-lengths of Furocoumarins : Psoralens

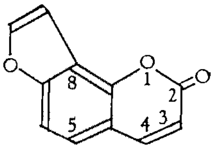
Biological photosensitized reactions are known to result from the absorption of a wide spectrum of radiation. The energy and therefore the wave-length of light absorbed and the biological changes occurring in photosensitized reactions are obviously closely related. A fundamental law of photochemistry, the Grotthus–Draper law, states that only radiation absorbed by the reacting system is effective in producing chemical change. Thus the radiation producing the photosensitizing reaction (that is, the action spectrum) should correspond to the absorption spectrum of the photosensitizing agent.

The furocoumarins, more generally known as psoralens, have been shown to be highly active photosensitizing agents capable of inducing sunburn or erythema and augmenting the skin pigmentation[1-3]. Furthermore, these compounds inhibit enzymatic activities as well as exhibit bactericidal and fungicidal effects following ultra-violet irradiation[4-7]. Their effect on ultra-violet carcinogenesis in albino and pigmented mice has also been investigated[8,9]. In most of these studies long-wave ultra-violet light has been observed by several investigators to be more effective in evoking these biological responses. The precise activating wave-length range was not known. In the course of examining the photosensitizing activity of 45 natural and synthetically prepared

Table 1. Activating and Fluorescent Wave-lengths of Furocoumarins and Related Compounds



Psoralen



*Iso*psoralen

No.	Compound	Activity erythemal response	Activating wave-lengths (mμ)	Fluorescence wave-lengths (mμ)
1.	Psoralen	Active	270, 360	435
2	4-methyl psoralen	Active	265, 315, 360	420
3	5′, 8-dimethyl psoralen	Active	270, 375	440
4.	4, 4′-dimethyl psoralen	Active	270, 360	435
5.	4, 5′, 8-trimethyl psoralen	Active	270, 370	440
6.	3, 4, 5′, 8-tetramethyl psoralen	Active	270, 360	430
7.	5′, 4, 8-trimethyl-3*n*-butyl psoralen	Active	275, 375	430
8.	8-methoxy psoralen (xanthotoxin)	Active	280 360	360 460
9.	5-methoxy psoralen (bergapten)	Active	270 280 350	370 485 400
10.	5′, 4-dimethyl-8-acetyl psoralen	Active	320, 370	430
11.	5′, 4-dimethyl-8-*n* propyl psoralen	Active	270, 360 400 450	430 460 525
12.	5′, 4-dimethyl-8-acetamido psoralen	Active	275, 360, 390	450
13.	5′, 4-dimethyl-8-acetyl-semicarbazone	Active	360	440
14	5′, 8-dimethyl-3, 4-benzo psoralen	Active	270, 300, 355	455
15.	5′, 8-dimethyl-3, 4-tetrahydrobenzo psoralen	Active	270, 360	430
16.	3-bromo-5′, 4-dimethyl psoralen	Active	275 375	430 460
17.	Marmesin (5′ isopropenol [2″]-4′, 5′-dihydro psoralen)	Active	270, 360	430
18.	Anhydro marmesin (5′ isopropyl-psoralen)	Active	270, 365	450
19.	5′ isopropenol [2″]-2 carboxy-4′, 5′-dihydro psoralen	Active	290, 340	380
20.	5′-methyl-isopsoralen	Active	280, 350, 400	460
21.	4′, 5′-dihydro-3-bromo-8-methoxy psoralen	Active	295, 380, 550	450
22.	5′, 4-dimethyl-8-amino psoralen	Inactive	380	530
23	4′, 5′-dihydro-4-methyl psoralen	Inactive	280, 360, 545	395
24.	4′, 5′-dihydro-8-methoxy-psoralen	Inactive	280, 365 450	480 525
25.	4′, 5′-dihydro-3, 5-dibromo-8-methoxy-psoralen	Inactive	280 290 300, 375	320 400 470
26.	5-bromo-8-methoxy psoralen	Inactive	280, 365 450	480 525
27.	5, 8-dimethoxy psoralen	Inactive	290, 360, 450	530
28.	4′, 5′-dihydro-5-ethylcarbamyl-8-methoxy psoralen	Inactive	290, 360 280	495 380
29.	8-hydroxypsoralen	Inactive	350	410
30.	4′, 5′-dihydro, 5′, 4-dimethyl-isopsoralen	Inactive	280, 355, 535	395
31.	2, 9-dimethyl-7H-furo-[3, 2-*f*-] [1]-benzopyran-7-one	Inactive	270, 360	435
32.	2, 9-dimethyl-7-H-oxazolo-[4, 5-*f*]-[1]-benzopyran-7-one	Inactive	265, 335 400 450	400 450 525
33.	benzoxazole-5-acrylic acid-6-hydroxy-β-7-dimethyl-γ-lactone	Inactive	350, 400	410, 470
34.	benzoxazole-5-acrylic acid-2, 6-dihydroxy-β-7-dimethyl-γ-lactone	Inactive	270 265, 380 310	470 490 420
35	benzoxazole-5-acrylic acid-6-hydroxy-β-2-7-trimethyl-γ-lactone	Inactive	270, 310, 355 400	400 470
36.	benzoxazole-5-acrylic acid-6-hydroxy 2-isopropyl-β-7-dimethyl-γ-lactone	Inactive	270, 310, 355	400
37.	2-carboxy-5, 6-dihydro-8-methoxy-benzo-(1, 2-b, 5, 4-b′)-difuran	Inactive	265, 335	430

Table 2. Activating and Fluorescent Wave-lengths of Furocoumarins

Compounds	Total	Activating λ (265–280 mμ)	Fluorescent λ (420–460 mμ)	Activating λ (340–380 mμ)	Fluorescent λ (420–460 mμ)
Biologically active*	21	15		20	
Biologically inactive*	16	2		2	

* As measured by erythemal response.

psoralens, it has been observed[10,11] that linearly annulated condensed tricyclic structure of psoralen exhibited optimum photosensitization on human as well as guinea pig skin.

Since fluorescence behaviour represents the emission from the activated molecules of absorbed light energy, we investigated the activating and the fluorescent wave-lengths of 37 furocoumarins and their biological photosensitizing action (Table 1). An Aminco–Bowman spectrophotofluorimeter was used in this study[12]. Each compound was dissolved in 95 per cent ethanol (1 mgm./10 ml.) and an aliquot used for determining the activation and fluorescent wave-lengths. The method for determining the biological photosensitization of furocoumarins and their relative activity as measured by erythemal response has been detailed elsewhere[10,11].

As indicated in Tables 1 and 2, the region of activating wave-lengths for photosensitizing action of furocoumarins lies between 265 and 280 mμ in the short ultra-violet range and between 340 and 380 mμ in the long-wave ultra-violet range. The fluorescence peaks for these activating wave-lengths were observed in the region of 420–460 mμ. The percentage fluorescence emitted was always significantly high when the activating wave-lengths were 340–380 mμ. Furocoumarins which induced definite photosensitized erythemal response on mammalian skin (human as well as guinea pig) showed this region of exciting wave-lengths. The inactive furocoumarins which did not exhibit photosensitizing response in general did not show these specific activating and fluorescent peaks. It appears that furocoumarins which show activation peaks in the region of 340–380 mμ and concomitantly the fluorescent peaks in the region of 420–460 mμ can cause photosensitization of skin in this region of long-wave ultra-violet light. The action spectra for these photosensitizing compounds therefore appears to be in the region of 340–380 mμ.

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[1] Fitzpatrick, T. B., Hopkins, O. E., Blickenstaff, D. D., and Swift, S., *J. Invest. Dermat.*, 25, 187 (1955).

[2] Psoralens and Radiant Energy; Proceedings of a Symposium, *J. Invest. Dermat.*, 32, 132 (1959).

[3] Musajo, L., Rodighiero, G., and Caporale, G., *Bull. Soc. Chim. Biol.*, 36, 1213 (1954).

[4] Pathak, M. A., and Fowlks, W. L. (in the press).

[5] Pathak, M. A., and Fellman, J. H. (to be published)

[6] Fowlks, W. L., Griffith, D. G., and Oginsky, E. L., *Nature*, 181, 571 (1958).

[7] Chakraborty, D. P., Das Gupta, A., and Bose, P. K., *Ann. Biochem. and Exp. Med.*, 17, 57 (1957).

[8] O'Neal, M. A., and Griffin, A. C., *Cancer Res.*, 17, 911 (1957).

[9] Pathak, M. A., Daniels, F., jun., Hopkins, C. E., and Fitzpatrick, T. B., *Nature*, 183, 728 (1959).

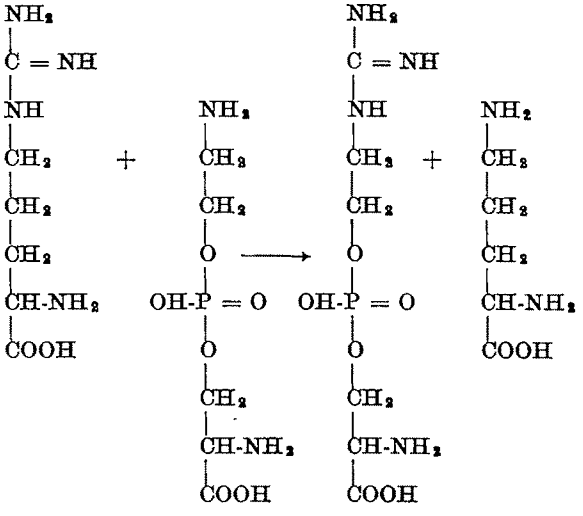
[10] Pathak, M. A., and Fitzpatrick, T. B., *J. Invest. Derm.*, 32, 509 (1959).

[11] Pathak, M. A., Fellman, J. H., and Kaufman, K. D. (in the press).

[12] Bowman, R. L., Caulfield, P. A., and Udenfriend, S., *Science*, 122, 32 (1955); 122, 972 (1955).

Biosynthesis of Lombricine

It was previously suggested[1,2] that serine ethanolamine phosphodiester (I) was the biological precursor of lombricine (II), a substance first isolated from earthworms by Thoai and Robin[3]. This suggestion was strengthened by the discovery of serine ethanolamine phosphodiester in earthworms[2,4] and by the finding that the serine moiety of both lombricine[5] and serine ethanolamine phosphodiester[4] had the unusual D-configuration. Experiments with inorganic phosphorus-32, [1 : 2-^{14}C] ethanolamine and DL-[3-^{14}C] serine have now provided additional evidence for this view. Furthermore, experiments with L-[amidino-^{14}C] arginine have shown that the amidino group of lombricine is derived from the amidino group of arginine (III), probably by a transamidinase catalysed transfer of the amidino group to serine ethanolamine phosphodiester with the formation of ornithine (IV):



(III) (I) (II) (IV)

The labelled precursors were administered to earthworms (*Megascolides cameroni*) in the amounts and specific radioactivities shown in Table 1. The labelled substances were given from a micro-syringe through a fine polythene tube inserted into the gastrointestinal tract of the worm for a distance of at least 3 cm. After the stated interval of time the worm was killed for the determination of the specific radioactivities of serine ethanolamine phosphodiester and lombricine (Table 1). The muscle was dissected out and extracted with 1·5 *N* perchloric acid. The extract was neutralized and then desalted on a 'Zeokarb 225' ion-exchange column (H$^+$ form). After concentration, the serine ethanolamine phosphodiester and lombricine were separated chromatographically on sheets of Whatman No. 3 filter paper with the solvents ethanol/formic acid/water (17 : 10 : 20 by vol.) and phenol/water (4 : 1 w/v). The specific radioactivities of the eluted serine ethanolamine phosphodiester and lombricine were determined using standard techniques. In some experiments the eluted lombricine was hydrolysed for 24 hr. at 110° in the presence of 6 *N* hydrochloric acid in a sealed tube, for determination of the distribution of radioactivity in the lombricine molecule (Table 2). The products of hydrolysis were separated chromatographically on paper strips with the phenol/water solvent. Serine from the serine moiety of lombricine and a combined area, representing guanidinoethanol

Table 1. *In vivo* INCORPORATION OF RADIOACTIVITY FROM LABELLED PRECURSORS INTO SERINE ETHANOLAMINE PHOSPHODIESTER AND LOMBRICINE OF EARTHWORM MUSCLE

Precursor	Amount administered (μmoles/gm.)	Radioactivity administered (c.p.m./gm.)	Time intervals (days)	Specific radioactivity (c.p.m./μmole)	
				Serine ethanolamine phosphodiester	Lombricine
Inorganic ^{32}P ..	carrier-free	$1{\cdot}1 \times 10^{6}$	2	560	20
Inorganic ^{32}P ..	carrier-free	$1{\cdot}1 \times 10^{6}$	4	840	97
[1 : 2-^{14}C] ethanolamine	3·7	$7{\cdot}5 \times 10^{6}$	4	67,900	1080
DL-[3-^{14}C] serine	2	10^{7}	2	31,500	904
L-[*amidino*-^{14}C] arginine	40	10^{6}	2	negligible*	329

* Net counting rate less than one-half the background counting rate.

Table 2 DISTRIBUTION OF RADIOACTIVITY IN LABELLED LOMBRICINE OF EARTHWORM MUSCLE

Precursor	Counts in serine moiety (per cent of total)	Counts in guanidinoethanol moiety (per cent of total)
[1 : 2-^{14}C] ethanolamine	6	94
DL-[3-^{14}C] serine	76	24
L-[*amidino*-^{14}C] arginine	negligible*	97

* Net counting rate less than one-half the background counting rate.

and guanidinoethyl phosphate from the guanidinoethanol moiety, were eluted for further counting.

After the administration of inorganic phosphorus-32, [^{14}C] ethanolamine and [^{14}C] serine, the specific radioactivity of the serine ethanolamine phosphodiester was much greater than that of the lombricine (Table 1), which is consistent with the view that serine ethanolamine phosphodiester is a precursor of lombricine and not a degradation product of that molecule. Table 2 shows that with [^{14}C] ethanolamine almost all the radioactivity was in the guanidinoethanol portion of the lombricine molecule, whereas with [^{14}C] serine most of the radioactivity was in the serine portion.

In contrast to the findings for inorganic ^{32}P, [^{14}C] ethanolamine and [^{14}C] serine, when L-[amidino-^{14}C] arginine was administered to the earthworm there was negligible radioactivity in the serine ethanolamine phosphodiester and considerable activity in the lombricine (Table 1). Almost all the radioactivity was in the guanidinoethanol moiety of the lombricine, indicating that at least one carbon atom of that portion of the molecule was derived from the carbon atom of the amidino group of arginine.

These findings strongly suggest that in the earthworm under *in vivo* conditions serine ethanolamine phosphodiester is the precursor of lombricine and that the amidino group of lombricine is derived from arginine by a transamidinase catalysed reaction.

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[1] Ennor, A H., and Morrison, J F, *Physiol. Rev.*, **38**, 631 (1958).

[2] Rosenberg, H, and Ennor, A. H., *Biochem. J.* (in the press).

[3] Thoai, N. v., and Robin, Y., *Biochim. Biophys. Acta*, **14**, 76 (1954).

[4] Ennor, A. H., Rosenberg, H., Rossiter, R. J., Beatty, I. M., and Gaffney, T., *Biochem. J.* (in the press)

[5] Beatty, I. M., Magrath, D. I., and Ennor, A. H., *Nature*, **183**, 591 (1959).

Dependence of Periodic Activation and Inhibition of Monoaminoxidase by Aliphatic Compounds upon Chain-length

MONOAMINOXIDASE has been linked with vascular and mental activity[1-3]. Among the most ancient modifiers of mental status are the alcohols, which have been shown[4] to modify monoaminoxidase *in vitro*. During our endeavours to develop this correlation further, we encountered a puzzling periodic activation and inhibition of this enzyme. For five years, the postulation of a tenable explanation has escaped not only our own interpretative capacities, but also those of the recognized specialists whom we have consulted. The recent studies of aliphatic compounds which have accompanied the discovery of irin[5] have provided our experiments with added interest: it now seems possible that enzymatic modification by aliphatic compounds might constitute a regulatory mechanism in the contraction of smooth muscle.

The determinations of monoaminoxidase activity were performed on rat liver homogenates and on isolated mitochondria with analytical methods previously discussed[6]. Among them the most reliable one[7] is the simple measurement of ammonia evolution[8]. Therefore only the results obtained with that method are shown here, although respirometry yielded qualitatively similar results whenever concomitantly performed.

The modification of the aminoxidase activity in a liver homogenate as a function of the chain-length of normal aliphatic alcohols is shown in Fig. 1. The alcohols had been subjected to elemental analysis which showed the absence of significant contaminants. The molarity of the alcoholic additives was $1{\cdot}5 \times 10^{-6}M$ for all the samples; the controls had saline added. The substrates were the hydrochlorides of tyramine and 1-norepinephrine (50 μmoles per ml. of reaction mixture). Short pre-incubations of the samples with the alcohols alone did not materially affect the results. The incubations were carried out at 4°C. since the modification was less pronounced at 37°C. and was diminished even more by higher temperatures. The results are expressed as percentages of the average reaction velocity of the control. The tyramine experiment is represented on the left ordinate and the norepinephrine results on the right. It is evident that the maxima and minima differ between the two substrates.

We have shown earlier that monoaminoxidase is intimately attached to the mitochondria of the rat liver cells[9] and that it may be rendered water-soluble by the use of suitable detergents[10]. At present, the most convenient detergent seems to be 'Igepal CO-630'[11]. As reported previously[10], the solubilization of this enzyme is accompanied by a significant increase in measurable activity. Thus it was of interest to determine whether the periodic phenomenon here discussed depended on the existence of intact mitochondria or whether the water-soluble enzyme-detergent complex also was capable of activation-inhibition with alcohols. Results with solubilized enzyme are summarized in Fig. 2 in the fashion discussed above. The normal alcohol concentration was $1{\cdot}8 \times 10^{-5} M$. It is noteworthy that the activation phase was evident only with norepinephrine and that with this substrate the maxima and minima were shifted as a result of the enzyme's solubilization.

Long ago, Blaschko[12] published a summary of his experience with the oxidation of aliphatic diamines of various chain-lengths by monoaminoxidase preparations. We interpret this activity curve as being periodic. Furthermore, what we consider as being the maxima in Blaschko's curve correspond to the minima in our tyramine curve, while his minima correspond to our maxima. Thus the two are essentially enantiograms of each other. It seems, therefore, that the phenomenon which we have encountered with alcohol additives might also be occurring when a homologous series of substrates are tested without any additive. It is also pertinent that the enzyme under discussion is sensitive to the substrate concentration, since we have described earlier[6] a marked temperature-dependent inhibition caused by high substrate concentrations. It was of interest therefore to study permutations of homologous series of substrates and of alcohols in order to discover the nature of the complementarity displayed by this system. As yet we have been unable to secure a homologous series of substrates in sufficiently pure form for enzymatic work.

In an effort to discover other aliphatic modifiers, we have investigated several series of aliphatic compounds. Of these the normal hydrocarbons have shown a behaviour qualitatively similar to the alcohols: a progressively increasing inhibition of the aminoxidase reaction was seen when the gaseous hydrocarbons were added to the atmospheric oxygen in equimolar concentrations. With the liquid hydrocarbons the modification of the enzyme occurred only in some of the experiments. When it did occur, it was as pronounced as with the alcohols. We have not as yet discovered the variable which caused this all-or-none response of our preparations. The normal saturated fatty acids and their methyl esters were inactive in several experiments.

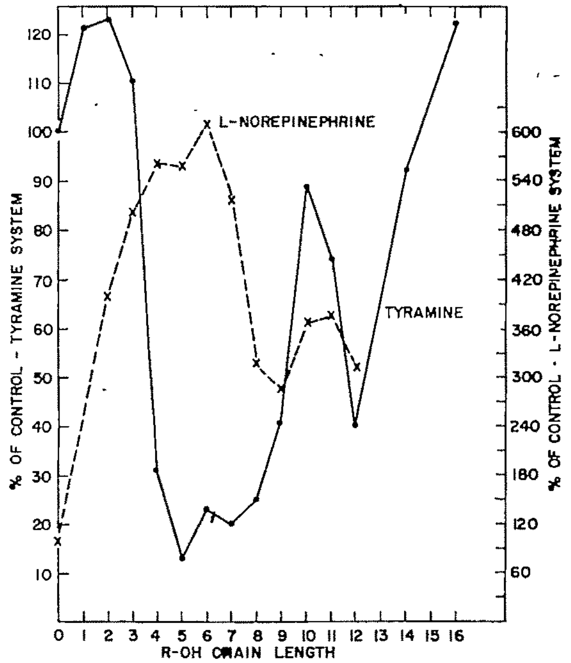

Fig. 1. Effect of chain-length of *n*-alcohols on monoaminoxidase activity of rat liver homogenate, as measured with the substrates indicated

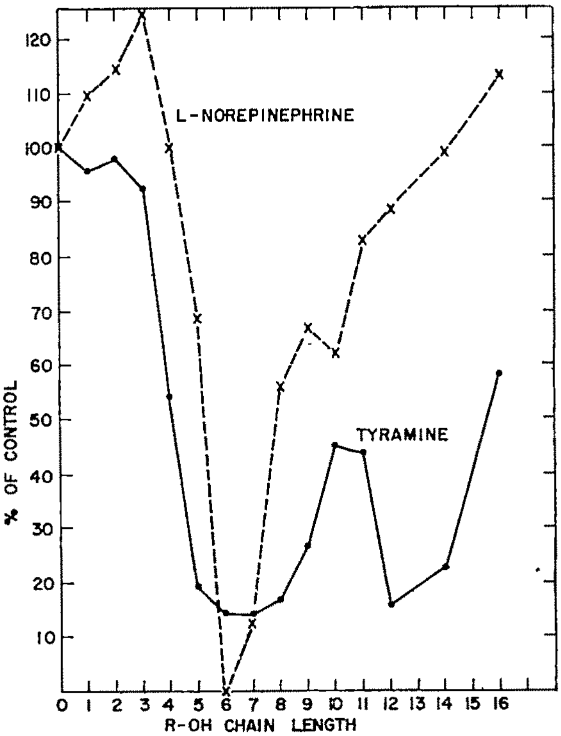

Fig. 2. Same as Fig. 1 but the homogenate was solubilized with detergent

It is evident that mammalian aminoxidase is amenable to periodic activation and inhibition by homologous series of some aliphatic compounds *in vitro*. The mechanism of this interesting phenomenon as well as its possible *in vivo* counterparts will require further work for their final definition.

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[1] Bogdanski, D. F., and Udenfriend, S., *J. Pharmacol. and Exp. Therap.*, 116, 7 (1956).

[2] Broadie, B. B., Pletscher, A., and Shore, P. A., *J. Pharmacol. and Exp. Therap.*, 116, 9 (1956).

[3] Broadie, B. B., Tourich, E. G., Kuntzman, R., and Shore, P. A., *J. Pharmacol. and Exp. Therap.*, 119, 461 (1957).

[4] Heim, F., *Arch. Exp. Path. Pharmakol.*, 210, 16 (1950).

[5] Ambach, N., *J. Physiol.*, 146, 255 (1959).

[6] Cotzias, G. C., and Greenough, J. J., *Arch. Biochem. Biophys.*, 75, 15 (1958).

[7] Cotzias, G. C., and Greenough, J. J., *Nature*, 183, 1732 (1959).

[8] Cotzias, G. C., and Dole, V. P., *J. Biol. Chem.*, 190, 665 (1951).

[9] Cotzias, G. C., and Dole, V. P., *Proc. Soc. Exp. Biol. and Med.*, 78, 157 (1951).

[10] Cotzias, G. C., Serlin, I., and Greenough, J. J., *Science*, 120, 114 (1954).

[11] Product of Antara Chemicals, a division of General Aniline and Film Corp.

[12] Blaschko, H., *Pharmacol. Rev.*, 4, 415 (1952).

Effect of Nitroacridine 3663 on Synthesis of Deoxyribonucleic Acid and Multiplication of T_2 Phage

It is known that the different acridine derivatives are able to become bound to isolated nucleic acids[1]. The compounds are more strongly bound by the highly polymerized deoxyribonucleic acid than by the ribonucleic acid[1]. The results of studying the interactions with deoxyribonucleic acid and the strong antibacterial effects of nitroacridine[2] suggest that these compounds may be used as virus chemotherapeutic agents.

The purpose of this work was to obtain information about the mode of action of nitroacridines. We used *Escherichia coli* strain *B* and phage T_2, employing the standard phage methods. The nitroacridine 3663 used was described by Schnitzer[2].

Bacteria were grown at 37° C. to a concentration of 2×10^8 per ml. in 2,500 ml. M-9 medium, sedimented and infected with 5 phage per bacterium in a non-nutrient adsorption medium[3]. The infected cells were then re-sedimented and transferred to a warm M-9 medium at 2×10^8 cells per ml. (zero time) which contained various concentrations of the compound.

The phage yields were measured at time zero and at other times from 1-ml. samples, diluted into a lysing medium[4] for intracellular bacteriophage determinations.

Immediately after infection and at various times later, samples of 500 ml. were withdrawn from the culture flask and quickly poured into 50 ml. of 3 M trichloroacetic acid. The fractions of deoxyribonucleic acid of the 500-ml. samples were obtained by a modified Schmidt–Thannhauser procedure[5]. Small aliquots of the trichloroacetic acid extracts containing deoxyribonucleic acid were analysed for deoxyribose by the diphenylamine method. The bulk of the extracts of deoxyribonucleic acid were evaporated to dryness and then hydrolysed[6]. The hydrolysates were chromatographed[6] and the amounts of the 5-hydroxymethylcytosine were determined spectrophotometrically.

The growth of the *E. coli B* was measured in M-9 medium by the colony plate count and by the nephelometric method. The compound was added in various concentrations to the culture when the growth was in the logarithmic phase.

As shown in Fig. 1, nitroacridine 3663 inhibits the synthesis of infective T_2 phage similarly to proflavine[4]; at the same time it has no effect on the multiplication of the host cell. The inhibition of the phage deoxyribonucleic acid occurs only at higher acridine concentrations. The degree of inhibition agrees entirely with the host cell.

It is thus seen that the effect of nitroacridine 3663 depends on the concentration of the compound. At very low concentrations (0·05–0·15 μgm./ml.) it inhibits the synthesis of T_2 phage without inhibiting the multiplication of the host cell. To account for this phenomenon it appears that during the vegetative phase of phage development the protein and deoxyribonucleic acid parts of phage are synthesized separately by the host cell[7]. The compound is bound more strongly by the structure-free phage deoxyribonucleic acid than by the structural nucleoproteins of the host. The deoxyribonucleic acid-acridine compound formed cannot be organized into an infective phage. In other words, this compound is a decoupling agent of the synthesis of phage nucleoprotein.

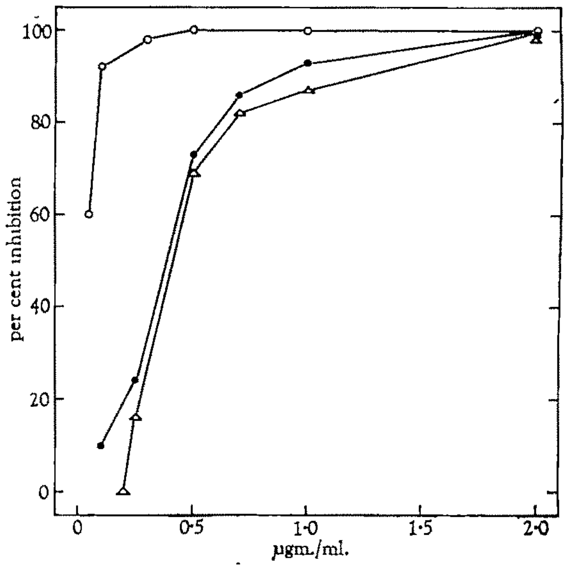

Fig. 1. Inhibitory effect of nitroacridine 3663 on —●—●—●— phage deoxyribonucleic acid synthesis (●); —○—○—○— multiplication of T_2 phage (○) and —△—△—△— multiplication of *E. coli B* (△), in per cent of untreated controls

At higher concentrations the compound inhibits the synthesis of phage deoxyribonucleic acid as well as the multiplication of *E. coli B*. The inhibitory effect is similar. From this observation it is concluded that the bacteriostatic effect of the compound is displayed by the inhibition of the synthesis of deoxyribonucleic acid.

Details of the relations of the structure and the effect of the different nitroacridines will be published elsewhere.

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[1] Irvin, J. L., and Irvin, E. M., *J. Biol. Chem.*, 206, 39 (1954).

[2] Schnitzer, R., and Silberstein, W., *Z. Hyg. Infektionskrankh.*, 109, 519 (1929).

[3] Hershey, A. D., and Chase, M., *J. Gen. Physiol.*, 36, 39 (1952).

[4] De Mars, R. I., *Virology*, 1, 83 (1955).

[5] Hershey, A. D., Dixon, J., and Chase, M., *J. Gen. Physiol.*, 36, 777 (1953).

[6] Vidaver, G. A., and Kozloff, L. M., *J. Biol. Chem.*, 225, 335 (1957).

[7] Hershey, A. D., The Harvey Lectures, 51, 229 (1957).

A Simple Test for the Presence of Metals in Biological Materials

The presence of metals in highly purified protein preparations is of considerable interest. The paramount part played by metals, especially in enzyme reactions (see, for example, ref. 1), is well known. Besides the specific metals in the metalloproteins, most proteins are able to bind, more or less firmly, metals removed from the medium. The removal of metals from a protein solution may be an important preliminary step in a study of its properties and structure. Unfortunately, the available methods for the detection of metals involve the use of very expensive equipment, or are laborious, and require large quantities of protein.

A very simple, non-specific test was devised to demonstrate the presence of metals in protein preparations. It is based on the spectral changes which occur when 8-hydroxyquinoline chelates metals. Spectral changes in the range of 300–370 mμ have already been studied[2]; but changes also occur at lower wave-lengths and have been used for this test. The alkali and alkaline earth metals give no response or a very weak one.

At neutral *p*H, 8-hydroxyquinoline has a very sharp absorption peak at 239 mμ which is shifted when chelation occurs to between 250 and 255 mμ, according to the metal involved (Fig. 1). The absorption here is approximately ten times that reported in the region of 360 mμ. It is fortunate for the effectiveness of the test that in this region of the spectrum most proteins have a minimal absorption.

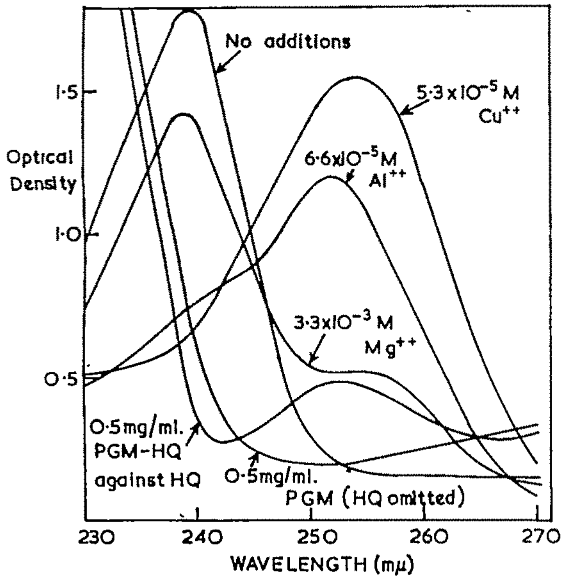

Fig. 1. Spectra of 8-hydroxyquinoline chelates. *Tris* *p*H 7·5, 8·3 millimolar; 8-hydroxyquinoline, 5·0 × 10^{-5} molar. Final volume 3·0 ml.

The general procedure for the test was as follows: The absorption of the protein (1 mgm. or less) in 1·0 ml. of 3·0 m*M* *tris*-chloride buffer *p*H 7·5 (the ionic strength was found not to be critical, stronger buffer can be used if necessary) was measured in a 'Unicam *SP* 500' spectrophotometer at 280 and 253 mμ against the same solution with protein omitted. To both blank and sample, 0·02 ml. of 2·5 m*M* 8-hydroxyquinoline was added and a reading at 253 mμ was made after mixing.

Precautions were taken to ensure that both blank and sample contained exactly the same constituents (except for the protein solution). The same pipettes were used and special care was taken to avoid contamination during mixing. Glass-distilled water was used, and was further de-ionized when very accurate determinations were required.

Results were expressed as:

$$\Delta E_{253/\text{mgm.}} = \frac{E' - E}{[\text{prot.}]}$$

where E is the optical density of the protein solution at 253 mμ, E' the optical density of the protein–8-hydroxyquinoline mixture minus the optical density of 8-hydroxyquinoline at 253 mμ, and [prot.] represents the protein concentration in mgm./ml. calculated from the E_{280} value.

Table 1. ULTRA-VIOLET ABSORPTION CHARACTERISTICS OF SOME METAL CHELATES OF HQ

Metals	Position of the peak mμ	Calculated absorption in presence of m*M* metal ions at 253 mμ
Ca^{++} and Ba^{++}	No peak observed.	—
Mg^{++}	255	0·125
Mn^{++}	253	1·170
Zn^{++}	255	25·000
Cu^{++}	254	57·200
Al^{+++}	251	17·200

HQ test as indicated in the text, but without protein. Sulphates of zinc, copper and aluminium, and the chlorides of the other metals were used.

In general the reaction was instantaneous; but in some cases the maximum absorption value was reached more slowly. The position of the peak varies slightly according to the metal as shown in Table 1, which also gives the calculated values of the optical densities for millimolar concentrations of some metals. Insolubility of the complexes was not found to be a problem, due to the very dilute solutions used, but at higher concentrations the optical density decreased after standing for a few minutes.

The test was applied to a crystalline phosphoglucomutase preparation[3], using between 0·3 and 0·8 mgm. protein per test. Phosphoglucomutase dialysed for 24 hours against large volumes of glass-distilled water gave a $\Delta E_{253/\text{mgm.}}$ value of 0·560 (Fig. 1). (From Table 1 a $\Delta E_{253/\text{mgm.}}$ value of 0·040 would correspond, for example, to about 0·06 mole of Cu^{++}, 0·2 mole of Zn^{++} or 2 mole of Mn^{++} per mole of enzyme (mol. wt. 74,000).) Passing this enzyme preparation through a carboxylic resin reduced the $\Delta E_{253/\text{mgm.}}$ to a negligible value, whereas dialysis against ethylenediaminetetraacetic acid, followed by exhaustive dialysis against glass-distilled water, increased this value.

The test was also used to follow the purification of phosvitin, which is known to contain iron, magnesium and calcium[5]. In this case the original $\Delta E_{253/\text{mgm.}}$ of 0·210 decreased to a negligible value after dialysis against ethylenediaminetetraacetic acid followed by extensive dialysis against water[6].

The test may also be applied to other materials provided that their absorption, at the wave-length used, is not too high. It proved very useful in showing metal contamination of one sample of glucose-1-phosphate which gave anomalous results when metal activation of phosphoglucomutase was studied. After passing the substrate through a sulphonic resin this anomaly was removed.

The 253 mμ peak was also used instead of the 360 mμ one in the determination of dissociation constants according to Burton[7] and for quantitative estimation of copper in pure solutions[4].

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[1] Vallee, B. L., "Adv. in Protein Chem.", 10, 318 (1955).
[2] Näsänen, R., *Acta Chem. Scand.*, 6 352 (1952). Näsänen, R., and Penttinen, U., *Acta Chem. Scand.*, 6, 837 (1952).
[3] Najjar, V. A., *J. Biol. Chem.*, 175, 281 (1948).
[4] (Unpublished observations.)
[5] Mecham, D. K., and Olcott, H. S., *J. Amer. Chem. Soc.*, 71, 3670 (1949).
[6] Williams, J. (personal communication).
[7] Burton, K., *Biochem. J.*, 71, 388 (1959).

ANIMAL PHYSIOLOGY

Comparative Adenylic Acid Deaminase Activity in Choroid Plexus and Ciliary Process Tissue

ANALOGIES between the ciliary process production of aqueous humour and the choroid plexus production of cerebrospinal fluid have been reviewed quite extensively by Davson[1]. The presence of an adenosine triphosphate-activated 5-adenylic acid deaminase in purified preparations of hog and rabbit ciliary process tissue[2,3] led to an interest in a similar system that might be present in choroid plexus tissue. It has, in fact, been shown by other investigators that 5-adenylic acid deaminase activity is present in preparations of whole dog brain[4,5].

Analytical methods employed in this investigation were as reported previously[2]. Incubations were done under the conditions described in Table 1.

Table 1. DEAMINASE ACTIVITY IN WHOLE-TISSUE HOMOGENATES
μMoles recovered after 40 min. incubation

Enzyme source	ATP	ADP	AMP	IMP	Adenosine	Total	μMoles NH_3
Dog choroid plexus	4 44	2·20	1 64	0 70	6·03	15 01	0·76
Hog ciliary processes	4 10	4·03	2 94	2·78	—	13 85	3·83

All vessels contained: 0 03 *M tris*-0 03 *M* succinate buffer, *p*H 7·4, 3 8 per cent fresh whole tissue homogenate; 7 μmoles each adenosine triphosphate and adenylic acid; 0 004 *M* magnesium chloride; 0·005 *M* potassium chloride; 0·025 *M* sodium chloride.

ATP, adenosine triphosphate; ADP, adenosine diphosphate; AMP, adenylic acid; IMP, inosinic acid.

Although the results indicate that choroid plexus tissue does have an active deaminase system, it is clear from the nucleotide recovery data that there are differences in the metabolic exchanges in the two types of tissue studied. We cannot as yet preclude the possibility that ammonia accumulation in the choroid plexus system could have resulted from deamination of any of the adenine derivatives recovered. The stoichiometry between the ammonia produced and inosinic acid accumulated is, however, strongly suggestive of the ammonia yield being the result of 5-adenylic acid deamination.

The fact that ammonia production in the ciliary process preparation was 5 times greater than in the choroid plexus preparation makes one question the activating influence of adenosine triphosphate as a deaminase co-factor in the latter tissue. This co-factor role of adenosine triphosphate has been reported for purified ciliary process preparations[6].

It has been shown previously that the deaminase of ciliary process is specific for 5-adenylic acid[2]. A similar specificity is seen in preparations of choroid plexus incubated under the same conditions described in Table 1 except substituting 14 μmoles of 3-adenylic acid as the nucleotide addition in some vessels and 14 μmoles of 5-adenylic acid as the nucleotide addition in other vessels. A yield of 0·2 μmole of ammonia was seen in the former case and 1·2 μmoles of ammonia in the latter—a 6-fold increase in activity with 5-adenylic acid.

Precise functional evaluation of the roles of these deaminase systems as regards actual aqueous humour and cerebrospinal fluid production must await further investigation. A role of adenine nucleotides in regulation of blood vessel patency[7,8] and roles of adenylic acid in many areas of metabolic interactions[9], either of which could invoke the need of an adenylic acid concentration-regulating deaminase, have already been suggested by others.

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[1] Davson, H., "Physiology of the Ocular and Cerebrospinal Fluids" (Little, Brown and Co., Boston, 1956).

[2] Waitzman, M. B., and Ballintine, E. J., *Amer. J. Ophthal.*, 46, No. 1, Part II, 96 (1958).

[3] Waitzman, M. B., *Fed. Proc*, 18, 165 (1959).

[4] Muntz, J. A., *J. Biol Chem.*, 201, 221 (1953).

[5] Mendicino, J., and Muntz, J. A., *J Biol. Chem*, 233, 178 (1958).

[6] Waitzman, M. B., *Amer. J. Physiol.* (in the press).

[7] Kutscher, W., and Sarreither, W., *Klin. Wchschr.*, 26, 698 (1948).

[8] Green, H., and Stoner, H., "Biological Action of the Adenine Nucleotides" (Lewis Publishing Co., London, 1950).

[9] Colowick, S. P., and Kaplan, N. O., editors, "Methods in Enzymology", 1, 197, 365, 585, 713, 740 (Academic Press, Inc., New York, 1955).

Effect of Kinetin on Glucose Metabolism of Chicken Embryo Fibroblasts

KINETIN, a potent plant-growth hormone, was recently discovered and identified as 6-(furfurylamino) purine, by Miller and Skoog[1]. Orr and McSwain[2] found that kinetin, at rather low levels (0·015–0·06 μgm./ml.), stimulated growth of human tissue culture.

In the course of a study of the effect of various hormones on cultures of chick embryo fibroblasts[3,4], it was observed that graded amounts of kinetin within the range of 12–24 μgm./ml. produced a roughly linear increase of total glucose consumption and lactic acid production (Fig. 1).

The ratio between glucose and lactic acid was similar in hormone-treated and control cultures. A kinetin concentration which stimulated glucose metabolism caused only slight growth inhibition. Microscopic examination revealed no abnormalities in the morphological appearance of the cells grown in the presence of the hormone.

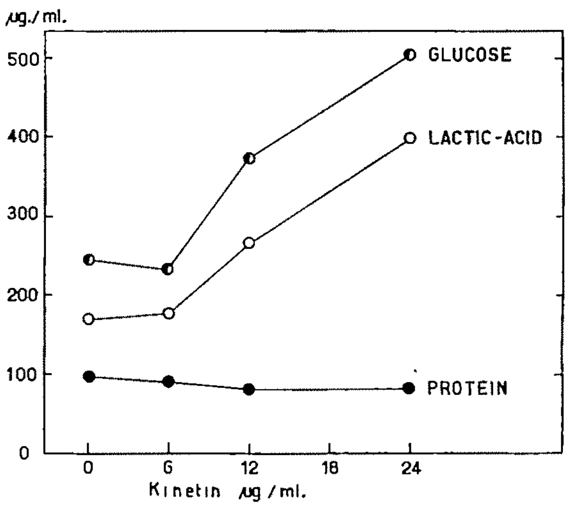

Fig. 1. The effect of kinetin on growth and glucose uptake of chick embryo fibroblasts and their lactic acid production. Procedures for cell cultivation and analytical methods were similar to those previously described (refs. 3 and 4). Cultures were incubated with hormone for 3 days prior to examination

In order to test the specificity of kinetin, a number of purines and pyrimidines and their derivatives were examined. Adenine and guanine showed a moderate and rather variable and irregular degree of stimulation; adenosine, guanosine, hypoxanthine, xanthine, cytosine, uracil and thymine did not affect glucose metabolism at a concentration up to 25 μgm./ml. Indoleacetic acid, another plant growth hormone known to affect the action of kinetin in plant tissues[5], did not modify the response of chick fibroblasts to kinetin, at a concentration up to 25 μgm./ml.

From the available data no conclusion can be drawn as to whether or not the described effect bears any relationship to the growth-promoting activity of kinetin in plants. The striking disparity between the effective levels in the two systems would rather indicate that the two phenomena are of an essentially different nature. It is not unlikely, however, that the quantitative difference is due to permeability factors.

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[1] Miller, C O., and Skoog, F., *J. Amer. Chem Soc.*, 78, 1375 (1956).
[2] Orr, M. F., and McSwain, B., *Cancer*, 10, 617 (1957).
[3] Halevy, S., and Avivi, L , *Biochim. Biophys. Acta*, 30, 198 (1958).
[4] Halevy, S., and Avivi, L. (in the press).
[5] Skoog, F , and Miller, C. O , *Symp. Soc. for Exp. Biol* , 11, 118 (1957).

Urinary Hyaluronidase

Ginetzinsky recently described a relationship between urinary hyaluronidase and urine concentration[1]. He found that urinary hyaluronidase assayed by his method was high at low urine volumes, virtually disappearing during water diuresis; but it remained high during an osmotic diuresis. From these results he inferred that antidiuretic hormone might act by stimulating the production of hyaluronidase within the nephron; this would facilitate water re-absorption by allowing water to pass through the ground substance between the cells. In an attempt to confirm Ginetzinsky's findings it became clear that his method[2] is unsuitable for quantitative assay of urinary hyaluronidase for the following reasons:

(1) Precise estimation of hyaluronidase by any viscosimetric method is dependent on a constant electrolyte composition of the hyaluronidase–hyaluronic acid system, as was demonstrated in 1940 by Madinaveitia and Quibell[3], and by McLean and Hale[4]. Urine has a variable electrolytic composition, and as Ginetzinsky does not appear to make any correction for this, his results are not quantitative.

(2) Ginetzinsky[1] fails to express his results as hyaluronidase excreted per minute, relying solely on arbitrary units per ml. If hyaluronidase were excreted at a constant rate like creatinine, then a graph of its concentration against urine flow per minute would be similar to Ginetzinsky's Fig. 1.

(3) He measures the initial viscosity of the mixture of urine, buffer and hyaluronic acid, the measurement being repeated after twenty minutes incubation, and the per cent reduction in viscosity is expressed as arbitrary units. This procedure is open to error as there is a rapid fall in viscosity before the initial reading[3] if a high concentration of hyaluronidase is present.

The method has been modified to allow for these factors. A detailed account will appear elsewhere, together with the results; but in principle the following changes have been made in the method of McLean and Hale[4]:

(1) The electrical conductivity of the urine is determined and compared with sodium chloride standards, the conductivity being brought to a value equal to that of 0·1 *M* sodium chloride in the final system by addition of a mixture of molar sodium and potassium chlorides.

(2) A micro-Ostwald viscosimeter is calibrated with a standard hyaluronidase preparation, incubation being at 37°C. The hyaluronidase concentration of the urine is obtained by reference to the calibration curve, and converted to units excreted per minute.

The experimental conditions chosen were water diuresis and mannitol osmotic diuresis as employed by Ginetzinsky: in addition, observations were made during potassium diuresis.

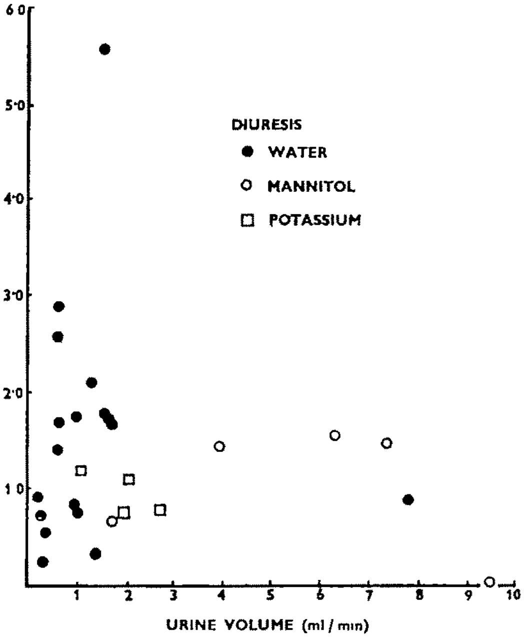

Fig. 1. Relationship of hyaluronidase output per minute to urine volume. There is no correlation between hyaluronidase excretion and urine minute volume in water diuresis or deprivation;

The results obtained demonstrate that first there is no relationship between hyaluronidase output (units per minute) and urine flow (ml. per min.) (Fig. 1). Secondly, there is in conditions of water deprivation and diuresis only an apparent relationship between hyaluronidase output and urinary osmolar excretion, sodium excretion and potassium excretion, the last three being interrelated. In osmotic diuresis in man with 25 per cent mannitol, hyaluronidase excretion remained at a fairly constant low level, in contrast to the findings of Ginetzinsky. If hyaluronidase concentration is plotted against urine minute volume (see Ginetzinsky's Fig. 2, ref. 2) there would naturally be a decrease in concentration as urine flow increases. Hyaluronidase excretion and potassium excretion both remain constant and are not related to sodium or osmolar excretion in an osmotic diuresis. In view of the constancy of potassium and hyaluronidase output in osmotic diuresis, the effect of a potassium diuresis on hyaluronidase output was observed. However, it was found that despite a rise in potassium excretion to 320 µequiv. per minute, there was no corresponding increase in urinary hyaluronidase.

Thus it is concluded that the mechanism of hyaluronidase excretion per minute is not directly related either to urine volume or to the excretion of sodium, potassium or total osmotically active solute.

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[1] Ginetzinsky, A. G., *Nature*, **182**, 1218 (1958).

[2] Ginetzinsky, A. G. (personal communication).

[3] Madinaveitia, J., and Quibell, T. H. H., *Biochem. J.*, **34**, 625 (1940).

[4] McLean, D., and Hale, C. W., *Biochem. J.*, **35**, 169 (1941).

[5] Dalgaard-Mikkelsen, S., and Kvorning, S. A., *Acta Pharmacol.*, **4**, 169 (1948).

Effect of Antihistamines on Mitochondrial Swelling and Liver Injury

Gallagher, Gupta, Judah and Rees[1] showed that thioacetamide, a liver carcinogen, would cause liver necrosis in rats and that a variety of antihistamines would afford protection. It was concluded that thioacetamide acted at the cell surface, but its mode of action was obscure, especially as no *in vitro* effect could be obtained. It has now proved possible to show an *in vitro* action of this compound. Liver slices, cut in the conventional manner from the livers of normal rats of the Wistar strain, show a gradual loss of certain dehydrogenases when suspended in saline solutions. Thioacetamide greatly increases this loss. Malic dehydrogenase was chosen as a suitable test system. The loss of this enzyme is shown in Table 1, which illustrates a typical experiment in the untreated slice. The effect of thioacetamide at a concentration of 0·02 *M* is shown and it can also be shown at concentrations of 5×10^{-3} *M*. These concentrations are in reasonable accord with the doses employed by Gallagher *et al.*[1], that is, 200 mgm./kgm., when it is remembered that extracellular water is of the order of 16 per cent and assuming uniform distribution of the drug.

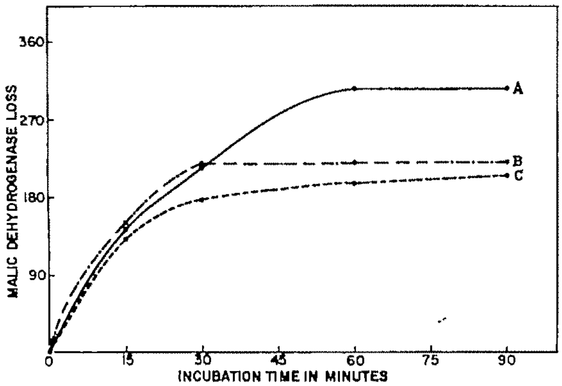

Fig. 1. Malic dehydrogenase activity was measured in the supernatant Ringer solution in which slices (about 150 mgm. wet wt.) were suspended. Samples were withdrawn as indicated and activity determined by following reduction of diphosphopyridine nucleotide spectrophotometrically. Temperature was 38°. Gas phase 95 per cent oxygen–5 per cent carbon dioxide. Dehydrogenase activity is shown in arbitrary units on the ordinates. Curve *A*, 0·02 *M* thioacetamide with calcium ion concentration 3×10^{-3} *M*; curve *B*, 0·02 *M* thioacetamide with calcium ion concentration 2×10^{-3} *M*; curve *C*, controls at both calcium-levels

The calcium-level of the medium is of importance in this action. Fig. 1 shows that reduction of the calcium-level from 3×10^{-3} *M* to 2×10^{-3} *M* results in a considerable diminution of the effect of thioacetamide. It will be seen that at both levels of calcium, the initial rates are the same but that after 30 min. the loss at the lower level is reduced to the control rate. The latter is relatively unaffected at either calcium-level. It may be inferred from this experiment that the effect of the calcium is concerned not with the initial attack of the thioacetamide, but with some subsequent step which is 'activated' by calcium, and which promotes the loss of intracellular components.

Table 1. Effect of Phenergan on Loss of Malic Dehydrogenase

Compound and concentration	Activity of malic dehydrogenase in medium as µmoles DPN reduced/100 mgm./hr. after 90 min. incubation
Nil	28·0
Thioacetamide, 0 02 *M*	40 0
Phenergan 10^{-4} *M*	24·0
Thioacetamide 0 02 *M* and phenergan 10^{-4} *M*	22 0

Liver slices (about 120 mgm. wet weight) were suspended in Krebs–Ringer, 5 0 ml., containing 0 01 *M* sodium bicarbonate with gas phase 95 per cent oxygen–5 per cent carbon dioxide. Temp. 38°. Total malic dehydrogenase activity of liver was 500 µmoles diphosphopyridine nucleotide reduced/100 mgm. liver/hr.

This interpretation is borne out by the experiments also shown in Table 1, where it will be seen that the phenothiazine antihistamine, phenergan, abolishes the effect of thioacetamide at a concentration of 10^{-4} *M*. The closely related compound chlorpromazine fails to protect these systems. Dawkins, Judah and Rees[2] have shown that phenergan prevents the accumulation of calcium in isolated perfused liver, and it may, therefore, be concluded that the action of phenergan on thioacetamide-induced leakage of malic dehydrogenase is not upon the initial attack of the drug, but upon the subsequent, calcium-'activated' step. This may also be inferred from the fact that phenergan at 10^{-4} *M* does not influence the normal rate of leakage of malic dehydrogenase from liver slices, a rate which is also uninfluenced by the calcium-levels tested, under the conditions of these experiments, but not necessarily under all conditions.

All the experiments cited above were performed in Ringer solutions buffered with sodium bicarbonate. If phosphate buffer is used at a concentration of 0·01 *M*, the action of thioacetamide on malic dehydrogenase is abolished. However, the initial attack of the drug may still be shown by following the leakage of total protein from the slices. Table 2 shows that even in phosphate buffer, loss of protein is accelerated by thioacetamide, and that this loss is unaffected by phenergan. Also shown in Table 2 is an experiment in which sodium bicarbonate is the buffer. It may be observed that a considerably greater loss of protein occurs in these circumstances and that phenergan exerts some effect. These

Table 2. Loss of Protein from Liver Slices

Compound and concentration	Mgm. protein in medium after 60 min. incubation	
	$NaHCO_3$	Phosphate buffer
Nil	5 0	3·6
Thioacetamide 0 02 *M*	8·2	5 8
Thioacetamide and phenergan 2×10^{-4} *M*	7 7	6 0
Thioacetamide and phenergan 4×10^{-4} *M*	7 3	—
Phenergan 2×10^{-4} *M*	6·3	3·5
Phenergan 4×10^{-4} *M*	6·[illegible]	—

The conditions were as in Table 1 for the experiments in sodium bicarbonate buffer; in the other experiments, phosphate buffer was added to 0·01 *M*, *p*H 7 2.

Table 3. ANTIHISTAMINES AND MITOCHONDRIAL SWELLINGS

System	Change in optical density in: 5 min.	10 min.	15 min.
Control	0·06	0·14	0·19
Phenergan 10^{-4} *M*	Nil	Nil	0·01
Benadryl 10^{-4} *M*	Nil	Nil	Nil
Pyribenzamine 10^{-4} *M*	Nil	0·03	0·03
Hispril 10^{-4} *M*	Nil	Nil	Nil

Mitochondria were suspended in 5·0 ml. 0·27 *M* sucrose–0·02 *M tris* buffer *p*H 7·2. The suspension was such that the initial reading was approximately 0·6. The changes were followed at 520 mμ on a Zeiss *PMQ*II spectrophotometer.

experiments support the hypothesis that after an initial attack by thioacetamide some degradative system is set into action. This is apparently dependent upon calcium ions and is inhibited by phenergan.

The nature of the initial attack of thioacetamide is under investigation and will be reported upon in the future. The nature of the degradative system is indicated by experiments shown in Table 3 demonstrating that several antihistamine compounds inhibit swelling of isolated rat liver mitochondria. They also protect these particles against the action of calcium ions, of hypotonic solutions and of thyroxine. They do so in concentrations and conditions in which they have no effect on respiration or phosphorylation of rat liver mitochondria, and it seems clear that they inhibit some mechanism by which isolated mitochondria accumulate water. In view of the demonstration in this work that thioacetamide action on isolated liver slices is dependent upon calcium ions and that its effect is due to a primary attack followed by a degradative reaction, it is tempting to believe that the latter is in fact the swelling and disruption of the mitochondria and that this is responsible for the liver damage caused by thioacetamide *in vivo*, which is also prevented by antihistaminics. In support of this view, thioacetamide does not affect mitochondrial swelling in cell-free preparations.

The results might also indicate a general mechanism for the action of the antihistamine compounds: it could be that all permeability changes are dependent upon mitochondrial changes, and that it is these which are prevented by the drugs in any event. Histamine then would merely trigger off the change in susceptible cells, for which it is specific, as thioacetamide is for the liver.

The actions of the antihistamines will be reported in detail shortly.

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[1] Gallagher, C. H., Gupta, D. N., Judah, J. D., and Rees, K. R., *J. Path. Bact.*, **72**, 193 (1956).

[2] Dawkins, M. J. R., Judah, J. D., and Rees, K. R., *J. Path. Bact.*, **77**, 257 (1959).

[3] Judah, J. D., *Exp. Cell. Res.* (in the press).

Respiration of Decapitated Mosquitoes

THE study of the basal metabolism of mosquitoes is rendered difficult because outbursts of locomotory activity may increase the respiration many-fold. Flight increases the oxygen uptake of insects 50–100 times as compared to resting rates[1] and causes variations in respiration between different groups of mosquitoes of the same strain.

We tried to eliminate the part of respiration due to locomotory activity in females of *Aedes ægypti* L. by decapitation. Decapitated mosquitoes are entirely inactive (unless purposely stimulated), but are able to survive for more than 24 hr. if kept at room temperature at a very humid atmosphere. We also measured the respiration of female mosquitoes after removal of their halteres, as haltereless mosquitoes retain some of their activity, but flight is limited to very short periods.

The respiration of intact, haltereless and decapitated mosquitoes, was measured in a Warburg apparatus at 28° C., using 10 females per cup. Readings were taken every 15 min. for 2 hr.

The oxygen consumption of decapitated mosquitoes was close to that of intact insects and much higher than that of the haltereless ones (Table 1). This observation seemed rather surprising, since haltereless insects are more mobile than decapitated ones; we suspected that the wound created by decapitation may have increased the gas exchange. When, however, the cuts were sealed with silicone grease, the respiration of the decapitated insects did not change at all.

Table 1. OXYGEN UPTAKE OF *Aedes ægypti* FEMALES

	No. of mosquitoes used	Oxygen uptake (mm.^{3}/hr./mosquito)
Intact mosquitoes	90	13·1±1·7*
Haltereless mosquitoes	190	8·2±0·6
Decapitated mosquitoes	190	14·7±0·9

* Standard error of the mean.

The effect of decapitation on the oxygen consumption is even better demonstrated at 12° C. At this temperature the intact insects are as immobile as the decapitated ones, and oxygen uptakes were 0·96 and 1·84 mm.^{3}/hr./mosquito respectively. The respiration of decapitated mosquitoes is quite steady, whereas intact insects and to a lesser extent haltereless insects exhibit variations due to bursts of activity.

Only 4–5 hr. after decapitation, the respiration of the insects begins to decrease; at the end of 24 hr. it is roughly equal to the respiration of haltereless insects.

Mosquitoes which were given a blood-feed 24 hr. prior to decapitation showed a much higher oxygen uptake than decapitated insects kept previously on a sugar–water diet (17 mm.^{3} and 11·4 mm.^{3}/hr./mosquito, respectively). Mosquitoes kept for 16 hr. prior to decapitation on a diet of water without sugar showed the same rate of respiration as those kept on sugar–water. Decapitation had no influence on the respiratory quotient, which was always between 0·9 and 1·1. In inactive intact mosquitoes, the spiracles are closed most of the time. Within a minute after decapitation the average spiracle aperture increases. Decapitation did not disturb the rhythm of the opening and closing of the spiracles. In mosquitoes which were decapitated while their spiracles were thus pulsating, the pulsation continued at the same rhythm, but the spiracles did not close completely. There was no increase in the ventilatory movements.

The corpora allata, which are known to influence oxygen consumption[2,3], are not removed by decapitation, but an inhibitory centre located in the head may be responsible for the increased oxygen uptake.

This increase in oxygen uptake as a result of decapitation was not observed with other insects. Raffy[4] reported that the oxygen uptake of adult silkworms decreased after decapitation, and I observed a similar effect with *Musca vicina* (Macq.) (Table 2).

Table 2. OXYGEN UPTAKE BY *Musca vicina*

	Oxygen uptake (mm.^{3}/hr./fly)
Intact flies	59
Haltereless flies	28
Decapitated flies	14

Decapitated and intact mosquitoes respond similarly to a reduction of the oxygen tension. The experiments were again carried out in a Warburg apparatus. In all cases the total pressure was maintained at one atmosphere by replacing oxygen by nitrogen.

Both groups maintained a nearly constant uptake of oxygen even in an atmosphere containing only 3–4 per cent of oxygen. The regulation of oxygen uptake at lower oxygen tensions is achieved in mosquitoes by changes in the spiracular activity. The spiracles control oxygen uptake by staying open for longer periods, by general enlargement of the aperture, and by increasing the frequency of opening. All three phenomena were observed in decapitated mosquitoes.

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[1] Chadwick, L. E., in "Insect Physiology", 630, edit. by Roeder, K. D. (Wiley, 1953).

[2] Thomsen, E., *J. Exp. Biol.*, **26**, 137 (1949).

[3] Samuels, A., *Biol. Bull.*, **110**, 179 (1956).

[4] Raffy, A., *Ann. Physiol. Physiochim. Biol.*, **10**, 437 (1934).

A Method of controlling the Temperature of Insect Neurosecretory Cells *in situ*

Periplaneta americana, like many other animals, can maintain a 24-hr. rhythm of locomotor activity when in constant conditions of light and temperature. A secretion produced by the neurosecretory cells in the sub-œsophageal ganglion has been shown to act as a controlling factor in the regulation of the phases of this rhythm[1]. Chilling cockroaches at 3° C. for some hours causes the phases of the activity rhythm to be retarded, in relation to solar time, by the number of hours for which the temperature is lowered. A method has now been devised whereby the temperature of the neurosecretory cells of the sub-œsophageal ganglion can be maintained at 3° C., while the rest of the body remains at room temperature. Neurosecretion apparently ceases at 3° C., and the phases of the neurosecretory cycle are retarded by the number of hours for which the cells have been chilled.

Since many functions of insects are known to be affected by the activity of neurosecretory cells, this method of temperature control may be of interest to other workers.

The apparatus is shown in Fig. 1. A copper bar is cooled at one end by a small vessel filled with cold alcohol. At the other end is a hollow conical tip carrying the cooling point, a 0·4-mm. copper wire. This wire is part of a copper constantin measurement thermocouple and is coated with polystyrene. Within the hollow tip is a thermistor. By means of an electronic unit[2] the thermistor controls the current through the heater attached to the bar, and maintains the tip at a constant temperature which is higher than equilibrium value. The alcohol is held at a low temperature by thermal circulation from a sealed chamber immersed in an ice and salt mixture and contained in a lagged tank. The separation of this primary cooler from mechanical contact with the rest of the device allows the bar to be mounted on a manipulator, and the position of the tip to be adjusted with precision.

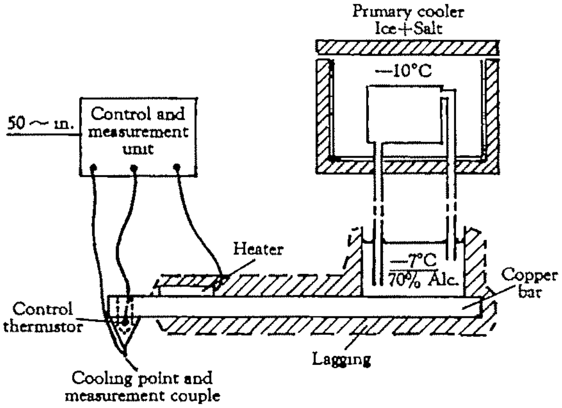

Fig. 1

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[1] Harker, J. E., *Biol. Rev.*, **33**, 1 (1958).

[2] Beament, J. W. L., and Machin, K. E., *J. Sci. Instr.*, **36**, 87 (1959).

Demonstration of Serum Precipitin to Brain Ganglioside

STUDIES on the structure of brain ganglioside, a macromolecular glycolipid which occurs in high concentration in cerebral grey matter, suggested that this material might be suited to function at membrane surfaces, since it possesses both carbohydrate and lipid moieties structurally so arranged that the molecule could bridge hydrophylic and lipophylic phases[1]. Further investigation of the properties of this substance[2,3] demonstrated that it interacted with certain viruses, both *in vitro* and *in vivo*, in a manner suggesting that it is utilized by infecting viruses as a natural receptor substance in the cell. In a membrane-active system, the clam heart, it was found to have stimulatory effect in very low concentration[4]. The presence of hexosamine and hexose constituents in brain ganglioside relates it structurally to the blood group substances[5]. For these reasons it became of interest to determine whether antisera might be prepared to brain ganglioside.

Since this preparation of brain ganglioside does not contain amino-acid or protein components, adjuvants might have been used in initial immunization attempts. However, it was decided to attempt to obtain an antibody response to the pure substance. Rabbits were injected at weekly and monthly intervals with 0·5 mgm. of bovine brain ganglioside salt in physiological saline, the first injections being made in the toe pads of the animal, and subsequent injections being made intravenously. The presence of antibody in rabbit serum was determined by collecting blood by free drip from the marginal vein of the ear,

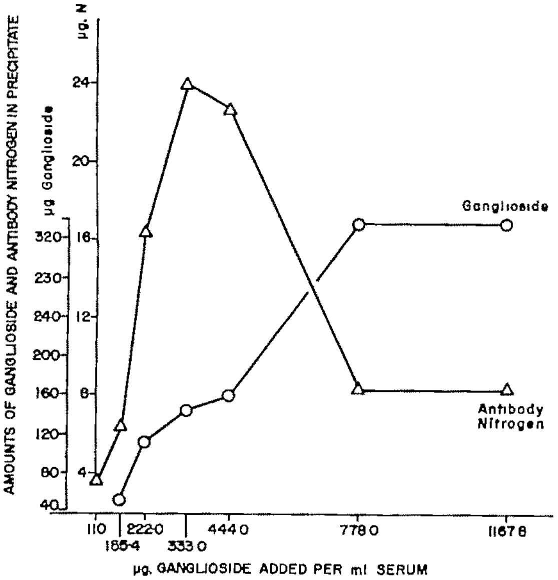

Fig. 1. Amounts of antibody nitrogen and brain ganglioside in the precipitate in the quantitative precipitin test

separating the serum, and determining the precipitin by means of the quantitative precipitin test described by Kabat[6]. The nitrogen in the precipitate was quantitatively determined by means of a Nessler's method[1]. Fig. 1 shows the results of a representative quantitative precipitin determination. In approximately one-half of the rabbits studied there was present a small amount of precipitin prior to immunization. The amount of precipitin in the serum could be increased by means of repeated injections of brain ganglioside. It may be noted that the peak of the precipitation curve is quite sharp. Because of the presence of neuraminic acid to the extent of 30 per cent by weight of the ganglioside, it was possible, by determining the neuraminic acid content, to determine the fate of ganglioside in both supernate and precipitate and thus to obtain some idea of the valence aspects of the precipitation phenomenon. The quantities in Fig. 1 are uncorrected for solubility of specific precipitate. In the zone of antibody excess, about 50 per cent of the added ganglioside is precipitated, suggesting that there is more than one immunologically distinct component in this preparation of brain ganglioside. With increasing amounts of ganglioside (into the zone of antigen excess) the precipitate contains decreasing amounts of antibody and increasing amounts of ganglioside. At the peak of the zone of maximum precipitation of antibody the precipitate contains approximately equal weights of brain ganglioside and antibody protein.

The antisera thus obtained have been used with the fluorescent antibody technique[7] to demonstrate the nerve cell body localization of brain ganglioside in bovine brain[10]. This method thus should be applicable to the histological localization of any normal or abnormal brain constituent to which antisera can be prepared.

The presence of a precipitin for brain ganglioside in mammalian serum, the titre of which is increased by successive injections of ganglioside, relates this normal brain constituent to other somatic or cellular antigens and blood-group substances immunologically as well as structurally. Other experiments in this laboratory[1-4] which have pointed to the likely membrane, transport, and receptor function of brain ganglioside are of interest in this regard. It may be that these antigenic and membrane roles are pertinently related in a manner which suggests the general function of the cellular substances which contain hexosamine or derivatives thereof (for example, muramic acid[8,9] and neuraminic acid). This function is here postulated to be the maintenance of the structural integrity and individuality of the specific cell in which they reside by the fact that their physico-chemical interaction with other molecules determines which enter and which leave the cell. Such function would have relevance to diverse biological phenomena.

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[1] Bogoch, S., *Biochem. J.*, 68, 319 (1958).
[2] Bogoch, S., *Virol.*, 4, 458 (1957).
[3] Bogoch, S., Lynch, P., and Levine, A. S., *Virol.*, 7, 159 (1959).
[4] Bogoch, S., and Bogoch, E. S., *Nature*, 183, 53 (1959).
[5] Kabat, E. A., "Blood Group Substances, their Chemistry and Immunochemistry" (Academic Press Inc., New York, 1956).
[6] *Ibid.*, 59.
[7] Coons, A. H., and Kaplan, M. H., *J. Exp. Med.*, 91, 1 (1950).
[8] Park, J. T., *J. Biol. Chem.*, 194, 877 (1952).
[9] Work, E., *Nature*, 179, 841 (1957).
[10] Bogoch, S., and Szulman, A. E. (to be published).

PLANT PHYSIOLOGY

Survival of the Twig of Woody Plants at −196° C.

I HAVE succeeded[1] in keeping alive living materials from the cell interiors of which all the easily freezable water was drawn by sufficient extra-cellular freezing at −30° C., after immersion in liquid nitrogen.

For the purpose of dehydration in various degrees, pieces from the same twig of a mulberry tree (2 cm. in length, 0·8 cm. in diameter) were pre-frozen for 16 hr. at 0°, −10°, −20°, −25°, *c.* −30° and *c.* −70° C. respectively and then immersed in liquid nitrogen. After being kept there for 10 min. they were held in air at −30° C. for 4 hr. and then transferred into air at 0° C. After this treatment the survival value of the parenchyma cells in the cortex of the twig was determined by the usual plasmolysis test. The percentage survival obtained in each case was as follows: 0 in the case of the pre-freezing at 0°, 10 at −10°, 40 to 60 at −20°, 60 to 70 at −25° and 100 at −30° to −70°. Further, in order to examine the the effect of repeated cooling, the same pieces of twig, after pre-freezing at −30° C., were immersed in liquid nitrogen for 10 min. and then were held in air at −30° C. for 1 hr. Such a process was repeated five times; but the living values were never lowered. These results indicate that if the twigs are previously frozen at −30° C., fatal intracellular freezing will never occur even at an extremely low temperature.

The same pieces of twig, after pre-freezing at −30° C., was held in liquid nitrogen for various periods of 10 min.–160 days, and the relation between the length of the immersing period and the proportion of survival was investigated. As a result of this experiment very long immersion in liquid nitrogen for 160 days at least was found to bring about no harmful effect on the plasmolysis capacity of the parenchyma cells of the twig. In addition, a twig of a willow (*Salix koriyanagi kimura*) and a piece of twig of mulberry tree in winter were not injured even when they were cooled down from room temperature to −90° C. under a cooling rate of 0·5° C./min. without pre-freezing and were maintained at −90° to −96° C. for 2 hr. It follows, therefore, that hardy material can be continuously cooled from room temperature to super-low temperatures without damage, if cooling is done at a slow rate.

Twigs of a mulberry tree pre-frozen at −30° C. gradually suffered damage as a whole after thawing from a super-low temperature, because their inner-cortex, pith-ray and pith-periclinal tissue are less resistant to freezing at −30° C. than are the parenchyma cells of the cortex. The twigs of poplar (*Populus siebodi* Mig.) and willow (*Salix koriyanagi kimura*) (15 cm. in length, 0·8 cm. in diameter), which are completely resistant to freezing at −30° C., had been kept in liquid nitrogen for 24 hr. After thawing, they were planted in moist sand in a greenhouse to test the capacity for development. The twigs so treated normally put forth buds and rooted. Most of the twigs pre-frozen at −20° C. put forth their buds; but they gradually withered within 30–50 days after planting. Twigs tested with liquid oxygen pre-frozen at higher temperatures than −15° C. never showed any sign of development. All the twigs of a willow pre-frozen at −30° C. revived even after they had been kept in liquid oxygen for 90 days. After being planted in a greenhouse in March 1959, their new shoots were about 90 cm. long by June 1 and are still growing normally.

In this pre-freezing method, the percentage survival is scarcely affected by the rate of cooling to and re-warming from a super-low temperature if the material is previously frozen at −30° C.[2]. If the living materials can withstand pre-freezing at a temperature lower than −30° C., they might be able to survive freezing at extremely low temperature by this pre-freezing method. It can be applied to materials with less frost hardiness, if any such anti-freeze agent as a glycerol is previously used on them so that freezing at −30° C. can be resisted.

Recently, Asahina[3-5] obtained some experimental results to support the view that this pre-freezing method is remarkably effective for keeping various animals alive in liquid oxygen. He kept a slug caterpillar[3], in overwintering prepupal stage, alive in liquid oxygen, the gill pieces of the oyster (glycerin used), overwintering larvæ of a butterfly, and a plant-parasitic nematode after pre-freezing at −30° C. Nei[6] kept yeast cells, suspended in distilled water, alive in liquid nitrogen, after a pre-freezing as low as −30° C.

From these facts it seems that there is a definite temperature at which almost all the easily freezable water in a cell may be drawn from the cell interior by extracellular freezing, and that the cells and tissues in this state are not injured even when exposed to an extremely low temperature. In the case of plants and animals this temperature lies around −30° C. Below this temperature, the intensity of cold seems not to exert any important effect upon woody plants, at least as long as the intense cold does not extend over a long period, and also it seems that the low temperature itself cannot be a primary factor of damage by frost.

Further details of this work will be published elsewhere.

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[1] Sakai, A., *Low Temp. Sci.*, Ser. B, **14**, 17 (1956).
[2] Sakai, A., *Low Temp. Sci.*, Ser. B. **16**, 41 (1958).
[3] Asahina, E., and Aoki, K., *Nature*, **182**, 327 (1958).
[4] Asahina, E., *Low Temp. Sci.*, Ser. B, **16**, 65 (1958).
[5] Asahina, E. (unpublished).
[6] Nei, T. (unpublished).

Effect of Direction of Growth on Sultana Canes

SULTANA vines are pruned to a limited number of one-year-old shoots (canes), each of which may carry up to 20 buds. As canes vary greatly in the proportion of fruitful buds the pruner aims to retain only the more fruitful ones on the vine. Such a selection is possible because the appearance of a cane indicates to some extent its fruiting potential[1]. But it is not known why canes of comparable origin differ both in appearance and in fruitfulness.

It was thought that the orientation of the growing cane in the gravitational field could be one contributing factor. Strong gravimorphic response has been reported for shoots on fruit trees[2]. Investigations were therefore carried out during 1958–59 to test the effect of gravity on sultana shoots, particularly with respect to the fruitfulness of their buds.

For 12 sultana vines, growing in the field at the Commonwealth Research Station, Merbein, the normal trellis height of 4 ft. was increased by erecting a vertical wire netting trellis to a total height of 7 ft. 6 in. Two shoots each in the centre of each vine were directed vertically upwards (0°), and at angles diverging from this direction by 45°, 90°, and 120°. The shoots were trained in their respective positions from October 16, more than one month before the initiation of the first bunch primordia, until the beginning of January, when the development of at least the first 20 buds was completed on the shoots attaining that length.

In June the length of all the shoots which had matured at least three buds was measured and the number of mature buds counted. All buds to position 20 were microscopically examined for the presence of bunch primordia which were extracted and weighed on an electro-balance. The results are shown in Table 1.

In fruit trees horizontal shoots grow less and form more flowers than vertical shoots[2]. This does not apply in the sultana where fruitfulness and bunch primordium development were severely depressed when canes were diverted from their normal direction of growth. Under natural conditions sultana shoots tend to grow vertically at least until flowering during the middle of November.

The total extension growth did not differ between the canes of the four treatments, but the canes of

Table 1. EFFECT OF DIRECTION OF GROWTH ON SULTANA CANES

	Canes deviating from the upward vertical direction by			
	0°	45°	90°	120°
No. of canes matured	16	16	9	15
Mean length of cane (cm.)	184·7	177·7	287·4	241·9
Mean internode length (cm.)	5·74	5·56	7·66	7·23*
Mean per cent fruitful buds†	39·7	44·7	17·3	20·4*
Mean bunch primordium weight (μgm.)	68·0	58·7	47·6	36·1*

* Treatments 0° and 45° differ significantly from treatments 90° and 120° in mean internode length, per cent fruitful buds, and mean bunch primordium weight.

† Statistically analysed as angular transformations.

treatments 90° and 120° had longer internodes. This could indicate that these canes were to some extent affected by being shaded for some hours daily by the rest of the vine. Heavy shading of sultana vines causes not only extension of internode length but it also reduces fruitfulness[3]. In treatments 90° and 120° bunch initiation and development may have been somewhat inhibited by the shading, but this was far too light to nullify any effect of gravimorphism such as found in fruit trees.

One could speculate that the grape vine differs from fruit trees and also from the Japanese larch[4] in its reaction to gravity because it is a climber under natural conditions. This problem may bear further investigation.

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[1] Antcliff, A. J., Webster, W. J., and May, P., *Aust. J. Agric. Res.*, 9, 328 (1958).

[2] Wareing, P. F., and Nasr, T., *Nature*, 182, 379 (1958).

[3] May, P., and Antcliff, A. J. (unpublished results).

[4] Longman, K. A., and Wareing, P. F., *Nature*, 182, 380 (1958).

Effect of Gibberellic Acid on Seeded Grapes

Size of grape berries was found by Muller-Thurgan in 1898 to be correlated with its number of seeds[1]. Size of seedless berries could be increased by means of auxins such as *p*-chlorophenoxyacetic acid[2,3] and by gibberellin spray[4,5]. The sprays had, however, no effect on size of seeded grapes[6,7].

Bunches of 'Queen of Vinyard' grapes were sprayed about 23 days after full bloom with 20 p.p.m. α-naphthalene acetic acid and gibberellin, respectively; 0·1 per cent 'Triton *X*-100' was added as wetting agent. 'Triton' alone was used for the controls. The fruit was harvested at full maturity, weighed, cut in halves and the number of seeds determined.

Results in Table 1 show a correlation of size of fruit and number of seeds. The 20 p.p.m. α-naphthalene acetic acid had no effect on fruit size; gibberellin, on the other hand, showed a marked effect. The added weight, however, decreased with the increasing number of seeds.

Table 1. Effect of Growth Substances on Grape Berries containing Different Numbers of Seeds. (Weight of 100 Berries in gm.)

Treatment	Seed No. per berry 0	1	2	3
Control	19	349	465	523
α-Naphthalene acetic acid	81	355	457	533
Gibberellin	181	491	506	526

This decrease is shown more clearly in the ratio between the weight of treated and untreated berries as affected by number of seeds (Fig. 1). The curve indicates that while the ratio is about 2·5 with parthenocarpic fruit it approaches 1 when the number of three seeds is reached.

It can be concluded, therefore, that gibberellin has a physiological effect on seeded berries, but only to a limited extent. Gibberellin compensates for the 'lack' of seeds; but once the berry has a sufficient number of seeds, gibberellin at the concentration used had no further effect.

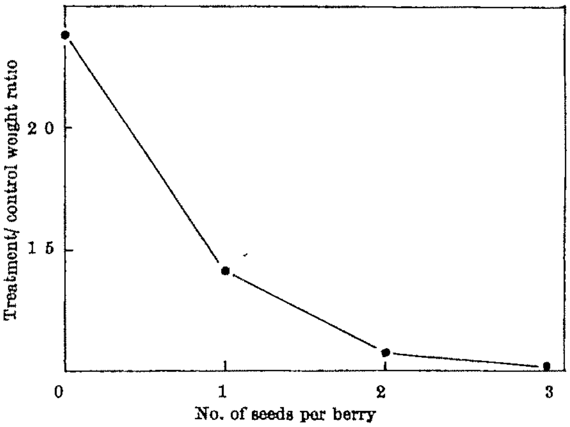

Fig. 1. Ratio between weight of gibberellin-treated and control grape berries as affected by number of seeds

No seed-like formations were found in the fruit due to gibberellin, though the tissue was more firm at the missing seed locations. α-Naphthalene acetic acid showed no effect.

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[1] Muller-Thurgan, M., "Land W. Jb. Schweiz", 135 (1898).

[2] Coombe, B. C., *South Austral. J. Agric.*, 57, 107 (1953).

[3] Weaver, R. J., *Proc. Amer. Soc. Hort. Sci.*, 61, 135 (1953).

[4] Stewart, W. S., Ghung, F. T., and Halsey, D. D., *Lasca Leaves*, 7, 80 (1957).

[5] Weaver, R. J., and McCune, S. B., *Hilgardia*, 27, 189 (1957).

[6] Samish, R. M., and Lavee, S., *Agric. Res. Sta. Rehovot 'Ktavim'*, 8, 273 (1958).

[7] Weaver, R. J., and Williams, W. O., *Bot. Gaz.*, 113, 75 (1951).

Mutagenic Action of Mitomycin C on *Escherichia coli*

Mitomycin *C* is of biological interest not only because it selectively inhibits cellular deoxyribonucleic acid synthesis[1,2] but also in that it induces lambda phage development in *Escherichia coli K* 12[3].

In this communication we present evidence that mitomycin *C* also affects the genetic mechanism, such as exerting a mutagenic action and stimulating the recombination rate in *Escherichia coli*.

Mutagenic action of mitomycin *C* was demonstrated in the reversion of *Escherichia coli B/r* tryptophan-requiring strain to the wild type. Logarithmic growing cells of this strain were harvested by centrifugation and resuspended in glucose-salt medium containing mitomycin *C* at a concentration of 1 μgm./ml. and incubated at 37° for 15 min. After the agent was removed by centrifugation, a part of the treated cell suspension was plated both on nutrient medium and minimal medium in order to count the total number of viable cells and the revertants to the wild type, immediately after the treatment. Another part of the treated cell suspension was incubated in broth after the treatment and assayed by the same method after one hour incubation.

During the treatment with mitomycin *C* the total number of viable cells decreased to one-tenth of the initial but during the post-incubation in broth the number of viable cells remained constant. Immediately after the treatment with mitomycin *C* the ratio of number of prototroph colonies to the total number

Table 1. EFFECT OF MITOMYCIN *C* ON THE YIELD OF THE WILD TYPE

Exp. No.	Conc. of mitomycin *C*	Time of incubation after the treatment 0	30 min	60 min.
1	1 μgm /ml.	36*	90	77
	0	1·2	1 5	0·3
2	1 μgm /ml.	25	182	177
	0	1 8	1·9	1·3

* Number of revertants per 10^8 viable cells.

of viable cells increased about tenfold of spontaneous reversion ratio. Much increase was found in the post-incubated cells (Table 1).

The effect of mitomycin *C* on the recombination rate of *Escherichia coli K* 12 was also examined. Cells of *Escherichia coli K* 12 58–161 *F*+ methionine-requiring strain were treated with mitomycin *C* in the same way as described above. The treated *F*+ cells were crossed with cells of *Y*–70 *F* – threonine, leucine and thiamine requiring strain, immediately after the treatment or 1 hr. incubation in broth followed by the treatment.

The ratio of prototrophic recombinants to the total viable cells was also increased five- to ten-fold and a greater enhancement was confirmed in the post-incubated sample. The reciprocal treatment of the recipient *F*– cells was ineffective to increase this ratio. The question remains as to whether the donor cells which lost their ability to form colony by the treatment with mitomycin *C* retain their capability to donate their chromosome.

These effects of mitomycin *C* on *Escherichia coli* obtained here were very similar to those due to ultra-violet radiation.

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[1] Shiba, S., Terawaki, A., Taguchi, T., and Kawamata, J., *Nature*, **183**, 1056 (1959).

[2] Sekiguchi, M., and Takagi, Y., *Nature*, **183**, 1134 (1959).

[3] Otsuji, N., Sekiguchi, M., Iijima, T., and Takagi, Y., *Nature*, **184**, 1079 (1959).

HÆMATOLOGY

In vitro Synthesis of Fœtal and Adult Hæmoglobin by Fœtal Hæmatopoietic Tissues

THE demonstration of a biochemical distinction between adult and fœtal forms of hæmoglobin (hæmoglobins *A* and *F*) has stimulated interest in their physiology and biochemistry[1]. Little is known about the site of origin or conditions governing the production of these two hæmoglobins. It has been postulated that hæmoglobin *A* is synthesized in the bone marrow and that hæmoglobin *F* is produced principally in the liver and spleen[2,3]. This theory corresponds well with the known sites of red cell production in the fœtus and in the adult[3]. As a corollary, it has been proposed that hæmoglobin *A* and hæmoglobin *F* occur in separate red cells[2,4]. Anoxia has been thought to increase the production of hæmoglobin *F*[4]. This report presents a study of the synthesis of hæmoglobins *A* and *F* by hæmatopoietic tissue from two white human fœtuses aged 17 and 9 weeks.

A viable fœtus of 17 weeks gestation (by menstrual history) was obtained immediately after surgical removal. Cell suspensions (approximately 100×10^6 nucleated cells per aliquot) were prepared from liver, spleen and bone marrow[5]. The cells were incubated for 24 hr. in 2 ml. of 50 per cent adult serum (type *AB*) in Hanks's solution (*p*H 7·6) containing 125 μgm. (2·7 μc.) of glycine-2-^{14}C. The percentages of oxygen in the gas phase during incubation are shown in Table 1. At the end of incubation the cells were washed three times with saline, and an excess of carrier hæmoglobin was added. The carrier contained 60 mgm. of hæmoglobin *A* and 60 mgm. of hæmoglobin *F*, prepared as a mixture of adult and cord blood. The alkali denaturation procedure[6] was used to determine the amount of hæmoglobin *F* in the cord blood. The samples were converted to carboxyhæmoglobin, and the adult and fœtal fractions were separated on an ion exchange column (1 × 10 cm.) using 'Amberlite *IRC*-50 (*XE* 64)' with a sodium citrate buffer at *p*H 6·5 as described by Prins and Huisman[7]. The separated hæmoglobins were dialysed for 48 hr. against distilled water, dried and counted at infinite thickness in a flow gas counter.

The results of the experiment are shown in Table 1. The measurements presented do not support the postulation of separate organ sites for the production of adult and fœtal hæmoglobins[2,3]. Relatively more hæmoglobin *F* was produced by cells from liver and spleen than from bone marrow, but the bone marrow of this fœtus produced twice as much hæmoglobin *F* as hæmoglobin *A*.

Anoxia did appear to induce a relative increase in the proportion of hæmoglobin *F* as compared to hæmoglobin *A*. However, it should be pointed out that the synthesis of both hæmoglobins decreased with decreasing oxygen tension.

Table 1 also shows a comparable experiment using the liver of a fœtus of 9 weeks gestation. Radioactivity indicating hæmoglobin synthesis was found both in the adult and in the fœtal fraction and in a proportion of about 1 part adult to 2 parts fœtal.

Walker and Turnbull[4] reported that hæmoglobin *A* is absent from fœtal blood until the 13th week of gestation. In the experiment shown in Table 1, hæmoglobin *A* was actively synthesized in the liver of the fœtus of 9 weeks gestation. Since synthesis of hæmoglobin *A* in hæmatopoietic cells must begin before significant amounts appear in the blood, the observations of Walker and Turnbull are not

Table 1. RADIOACTIVITY OF HÆMOGLOBIN EXPRESSED AS COUNTS PER MINUTE AT INFINITE THICKNESS

Exp.	Oxygen Per cent	Liver Hb-*A* c.p.m.	Liver Hb-*F* c.p.m.	Liver Hb-*A*/Hb-*F*	Spleen Hb-*A* c.p.m.	Spleen Hb-*F* c.p.m.	Spleen Hb-*A*/Hb-*F*	Bone marrow Hb-*A*	Bone marrow Hb-*F*	Bone marrow Hb-*A*/Hb-*F*
A	20 (air)	535	2600	1/4 8	47	189	1/4 0	465	870	1/1·8
	10	372	1950	1/5·3						
	5	101	816	1/8·0						
	0	29	55							
B	20 (air)	1280	2940	1/2 3						

Exp. A, 17 week fœtus, crown-rump measurement 13 cm.; Exp B, 9 week fœtus, crown-rump measurement 2·1 cm.

necessarily incompatible with those here reported. At a given time the ratio of synthesis of hæmoglobin *A* to hæmoglobin *F* must be greater than the ratio of these two hæmoglobins in the blood due to the life-span of the red cells, to the increasing percentage of hæmoglobin *A* and to the expanding blood volume of the fœtus. Should fœtal red cells containing hæmoglobin *A* have a relatively shorter life-span than those containing hæmoglobin *F*, the synthetic ratio, *A*/*F*, might be even higher. However, our observations do not bear upon the question of whether separate red cells contain exclusively one or the other types of hæmoglobin[3,4].

We have assumed the presence of only two hæmoglobins, *A* and *F*. It is now generally recognized that these two fractions are heterogeneous[8]. Experiments to characterize these hæmoglobin subfractions and to evaluate their importance in hæmoglobin synthesis are in progress.

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Aug. 31.

[1] White, J. C., and Beaven, G. H., *Brit. Med. Bull.*, 15, 33 (1959).

[2] Langley, F. A., *Arch. Dis. Childhood*, 26, 64 (1951).

[3] Brody, S., *Lancet*, 1, 520 (1959).

[4] Walker, J., and Turnbull, E. P. N., *Arch. Dis. Childhood*, 30, 111 (1955).

[5] Thomas, E. D., *Blood*, 10, 600 (1955).

[6] Jonxis, J. H. P., and Visser, H. K. A., *Amer. Med. Assoc. J. Dis. Children*, 92, 588 (1956).

[7] Prins, H. K., and Huisman, T. H. J., *J. Lab. and Clin. Med.*, 46, 255 (1955).

[8] Allen, D. W., Schroeder, W. A., and Balog, J., *J. Amer. Chem. Soc.*, 80, 1628 (1958).

Plasma Antihæmophilic-Factor Concentrations in the Australian Aborigine and in Conditions associated with Hyperglobulinæmia

While studying the plasma antihæmophilic-factor concentrations of a large number of healthy individuals, it was noted that Australian aborigines in general have higher plasma concentrations of this factor than the white Australian population. The factor was estimated by a technique[1] based on the thromboplastin generation test and results were expressed as a percentage of the concentration in a standard normal plasma. The lower histogram in Fig. 1 shows the plasma concentrations in 86 healthy white adults adjusted to the nearest 10 per cent. The values are skewed with a fairly sharply defined lower limit and range from 60 to 200 per cent of standard normal. This group comprised equal numbers of males and females, and there was no significant sex difference in the values. These normal results are very similar to those found in a previous group of 80 normal white adults reported on from this laboratory[2].

Concurrent with the examination of this normal group, blood samples were examined from 28 Australian aborigines who were hospital in-patients.

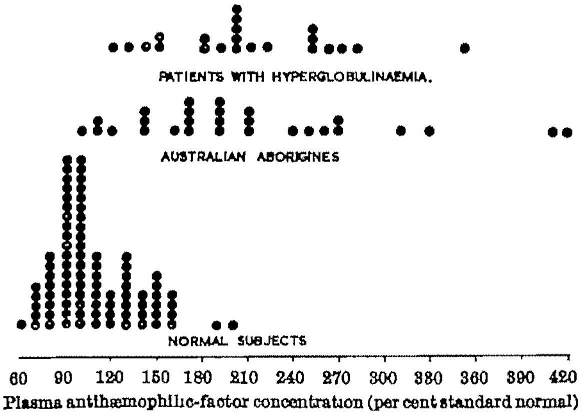

Fig. 1. Plasma antihæmophilic-factor concentrations in normal subjects, Australian aborigines and patients with hyperglobulinæmia

The plasma antihæmophilic-factor concentrations are shown in the middle histogram in Fig. 1. Values ranged from 100 to 420 per cent of standard normal and twelve samples showed values greater than the upper limit of the normal white range. The natives were of mixed purity from half caste to full blood. They were in hospital mostly for surgical or traumatic reasons. The higher values could be the result of different racial origin. However, the Australian aborigine has a higher concentration of plasma γ-globulin than the white Australian[3]. It was therefore decided to investigate white Australians who had hyperglobulinæmia as the result of disease.

The upper histogram in Fig. 1 shows the plasma antihæmophilic-factor concentrations in 22 white Australians who had hyperglobulinæmia as a result of varying diseases including lymphosarcoma, auto-immune disease, hepatic cirrhosis, nephrosis and multiple myelomatosis. Values ranged from 120 to 350 per cent of standard normal and approximated more to the aboriginal group than the white normal group. Nine samples showed values greater than the upper limit of the white normal range.

The plasma antihæmophilic-factor value is thought to be genetically determined and constant for the individual. The findings in the patients with hyperglobulinæmia indicate that the value can alter with disease. That plasma antihæmophilic-factor concentration may be related to the γ-globulin concentration is unexpected since the antihæmophilic-factor migrates electrophoretically with fibrinogen.

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[1] Pitney, W. R., *Brit. J. Hæmat.*, 2, 250 (1956).

[2] Pitney, W. R., and Arnold, B. J., *Brit. J. Hæmat.*, 5, 184 (1959).

[3] Curnow, D. H., *Med. J. Aust.*, ii, 608 (1957).

Absorption–Elution Grouping of Dried Blood Smears

Thick blood smears (5–10 μl./cm.^{2}) can be grouped in the ABO blood group system by an absorption-elution technique which has been evolved in this Laboratory over the past two years.

Dried blood smears, on standard microscope slides, prepared in the usual way, are fixed by plunging in *p*H 7·4 McIlvane buffer at 100° C. for 30 sec. The denatured smears are then blotted dry and divided into separate cavities bounded by gum dammar/beeswax mixture[1], the number of cavities being governed by the number of antisera to be employed. Antisera are then dispensed into the relevant cavities in excess of the amount required to saturate the antigen. The slide is allowed to stand at room temperature, in a moist chamber, for 3 hr. with occasional gentle agitation, then rinsed under the cold tap and blotted dry. Drops of 2 per cent v/v red cell suspension are next dispensed into the different cavities and the temperature of the slide is raised to 50° C. for 5 min. in a preheated moist chamber.

On removal to room temperature, and gentle rotation, agglutination of the red cells occurs in those cavities where antibodies have been eluted by the elevated temperature.

Typical results obtained are shown in Table 1.

Table 1

	Antiserum and cells added					
Group of blood smear	Anti-A A_1	Anti-A_1 A_1	Anti-B B	Group O A_1	Group O B	Gorse Anti-H O
A_1	+	+	−	+	±	−
A_2	+	−	−	+	±	+
B	−	−	+	±	+	±
O	−	−	−	−	−	+
A_1B	+	+	+	+	+	−
A_2B	+	−	+	+	+	+

+, Macroscopic agglutination; ±, variable results, −, no agglutination.

The agglutination is, in nearly all cases, visible to the unaided eye, although with some thinner blood smears microscopic examination may be necessary. Using group O antiserum, an A blood smear (especially if very thick) may elute anti-B and conversely a group B blood smear may elute anti-A. This is in agreement with the results obtained from antibody elution from intact cells[2]. Gorse extract is a source of potent and specific anti-H[3] while the anti-A_1 antiserum used is pooled group B absorbed with A_2 cells. The appearance of a slide bearing a smear of A_2 blood on completion of grouping by the above technique is shown in Fig. 1.

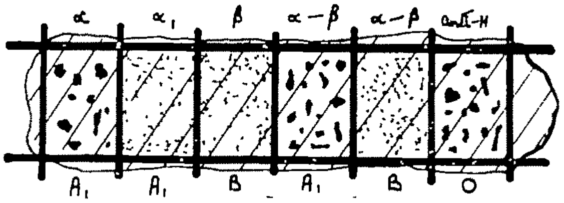

Fig. 1

The anti-A and anti-B sera used (cols. 1 and 3) have given unequivocal results in all tests we have made. The group O serum reactions (cols. 4 and 5) have given consistently weaker reactions than separate anti-A and anti-B, but we have included it as a check and as being of possible significance when testing for subgroups of A weaker than A_2. The anti-A_1 and anti-H reactions have in most cases given good reciprocal results.

The technique has obvious uses in the field of blood grouping, particularly since we have recently found that it can be applied to smears made from dried bloodstains. The advantages are that no antisera of standardized titre are required, no interference by detergents is encountered (these are extracted by the hot water) and the results can be read immediately without tedious pipetting and microscopic examination. The smears can be stored indefinitely and repeat grouping can simply be made by rinsing the cells and antibodies off the slide with water at 50° C. and starting again as with a freshly fixed blood smear.

The method shows obvious promise, and merits further investigations to an extent which is not possible in a routine laboratory. These preliminary observations are published in the hope that research workers in blood group serology will take the study further.

I am indebted to Dr. I. Dunsford for criticism, antisera and blood samples, and to Dr. I. M. Barclay, director of this Laboratory, for permission to publish this communication.

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[1] Dade, H. A., and Waller, S., Commonwealth Mycological Papers No. 27 (1949).

[2] Dodd, B. E., *Brit. J. Exp. Path.*, 33, 1 (1952).

[3] Boyd, W. C., and Shapleigh, E., *J. Lab. Clin. Med.*, 44, 235 (1954).

RADIOBIOLOGY

Evidence for Protein Turnover in an Embryonic System

IN a previous communication we have compared the relative efficiency of various proteins, peptides and free amino-acids as precursors of chick embryonic proteins[1]. From these studies we concluded that during the early stages of development the preferred precursor was yolk protein (rather than peptides or amino-acids) and that the pathway of synthesis of chick embryonic protein probably proceeded via a stage involving amino-acid derivatives (possibly amino acyl adenylates[2] or, more likely, amino-acids linked to the terminal adenosine residues of polyribonucleotides[3-6]).

In the present study we investigated the utilization by chick embryos of intravenously injected sulphur-labelled protein precursors in the formation of heart and liver, as well as the relative rates of growth of these organs, as indicated by the uptake of label by these organs.

Torulopsis utilis was grown on sulphate labelled with sulphur-35 according to the method of Wood and Perkinson[7]. After extraction with organic solvents[8], the yeast protein was hydrolysed with a 1:1 mixture of 6 *N* hydrochloric acid and 90 per cent formic acid. The humin was removed by centrifugation, and a portion of the neutralized hydrolysate corresponding to about 1 mc. of the original labelled sulphate per 30 gm. body-weight was injected intraperitoneally into a White Leghorn hen. In other experiments cystine labelled with sulphur-35 was removed from the yeast protein hydrolysate by adding carrier cystine, and precipitating this amino-acid at *p*H 4·5 in the cold overnight. This cystine-free yeast protein hydrolysate was then injected into baby chicks. Eight to twelve hours after injection the animals were bled by heart puncture, and serum albumin and serum albumin peptides were prepared by methods previously described[9]. Embryonated White Rock eggs were incubated at 38°C. under conditions of controlled humidity for a period of 9 days. Embryos were injected intravenously[10] with one of the following protein precursors on the ninth day of incubation: yeast protein hydrolysate containing either methionine labelled with sulphur-35 alone or containing both labelled methionine and labelled cystine; chicken serum albumin peptides containing sulphur-35 in the methionine residues or containing sulphur-35 in both the methionine and cystine residues; or chicken serum albumin containing labelled methionine. It should be pointed out that all preparations also contained some labelled methionine sulphoxide[9]. On the tenth day of incubation hearts and livers were excised[10], the organs pooled and homogenized, and dry protein powders prepared for counting[9]. The results of these experiments are shown in Table 1.

Table 1. DISTRIBUTION OF LABEL IN EMBRYONIC CHICK HEART AND LIVER ON THE TENTH DAY OF INCUBATION AFTER INTRAVENOUS INJECTION OF VARIOUS PROTEIN PRECURSORS ON THE NINTH DAY OF INCUBATION

Precursor injected	Per cent injected activity		Specific activity (Heart/Liver)
	Heart	Liver	
Yeast hydrolysate containing labelled methionine	0·62	1·61	1·30
	0·55	1·62	1·08
Yeast hydrolysate containing labelled methionine and labelled cystine	0·62	1·62	1·18
Chicken serum albumin peptides containing sulphur-35 in the methionine residues	0·55	1·66	1·23
	0·46	1·77	1·20
	0·56	1·48	1·37
Chicken serum albumin peptides containing sulphur-35 in the methionine and cystine residues	0·35	1·23	1·50
Chicken serum albumin containing labelled methionine	0·12	0·40	1·24
	0·29	0·56	1·95

The results are presented here in two ways: as the percentage of injected activity found in the two organs for each of the precursors injected, and as the ratio of specific radioactivity of heart/liver. The values indicating the percentage of injected activity are uniformly larger for liver than heart, as is expected on the basis of the difference in size of the two organs on the tenth day of development. It will be noticed, however, that all the values (independent of the type of protein precursor injected) for embryonic heart are quite close to one another, as are the values for embryonic liver.

These results indicate that while anabolism is certainly the predominant process in this rapidly proliferating system, some catabolic capacity must also be operative in order to account for the conversion of injected serum protein and peptides into embryonic tissues. This is in contrast to the finding that catabolism does not take place in rapidly growing bacterial systems[11,12], but corresponds more closely to the situation in resting bacterial suspensions where protein turnover is known to take place[13]. We had previously suggested[1] that in the conversion of yolk protein to embryonic chick protein the catabolic activity was centred in the yolk[14] while the synthetic activity resided in the embryo proper. The present results do not conflict with this conclusion but indicate that the embryonic system itself does contain a catabolic potential.

The ratio of specific radioactivity of heart/liver is also shown in Table 1 and is uniformly greater than 1·00. This indicates that both organs are capable of incorporating each of the various protein precursors tested at essentially similar rates; furthermore, over the period of the experiment the heart is anabolically more active than the liver (that is, within the time period tested a greater fraction of its newly synthesized protein has its origin in the precursors supplied).

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[1] Walter, H., and Mahler, H. R., *J. Biol. Chem.*, **230**, 241 (1958).
[2] Hoagland, M. D., Keller, E. B., and Zamecnik, P. C., *J. Biol. Chem.*, **218**, 345 (1956).
[3] Schweet, R. S., Bovard, F. C., Allen, E., and Glassman, F., *Proc. U.S. Nat. Acad. Sci.*, **44**, 173 (1958).
[4] Zachau, H. G., Acs, G., and Lipmann, F., *ibid.*, **44**, 885 (1958).
[5] Preiss, J., Berg, P., Ofengand, E. J., and Bergman, F. H., *ibid.*, **45**, 319 (1959).
[6] Hecht, L. I., Stephenson, M. C., and Zamecnik, P. C., *ibid.*, **45**, 505 (1959).
[7] Wood, J. L., and Perkinson, J. D., *J. Amer. Chem. Soc.*, **74**, 2444 (1952).
[8] Williams, R. B., and Dawson, R. M. C., *Biochem. J.*, **52**, 314 (1952).
[9] Walter, H., Haurowitz, F., Fleischer, S., Lietze, A., Cheng, H. F., Turner, J. E., and Friedberg, W., *J. Biol. Chem.*, **224**, 107 (1957).
[10] Walter, H., Allmann, D. W., and Mahler, H. R., *Science*, **124**, 1251 (1956).
[11] Spiegelman, S., in "Enzymes, Units of Biological Structure and Function", ed. by Gaebler, O. H. (Academic Press, 1956).
[12] Hogness, D. S., Cohn, M., and Monod, J., *Biochim. Biophys. Acta*, **16**, 99 (1955).
[13] Reviewed by Pollock, M. R., in "The Enzymes", 2nd ed. (ed. by P. D. Boyer, H. A. Lardy, and K. Myrback) (Academic Press, New York, 1959).
[14] Romanoff, A. L., *Ann. N.Y. Acad. Sci.*, **55**, 288 (1955).

Ear Tumours in Rabbits receiving Strontium-90

THE induction of bone tumours by radium and other 'bone-seeking' radioactive materials is now well known. It was originally observed in human cases of radium poisoning[1-4]. Experimentally, similar tumours have been produced in a wide range of animal species following the administration of either radium[5-9] or a variety of 'bone-seeking' radioactive isotopes such as strontium-89, strontium-90, plutonium-239 and calcium-45[10-19]. Tumours occur in the neighbourhood of sites of maximum isotope deposition and retention (see ref. 13). In young rabbits receiving a single injection of strontium-90[13], tumours occurred most frequently in the metaphysial regions of the long bones, while in both old and young rabbits[20] they occurred adjacent to growing teeth in the mandible and maxilla.

Less well known are the tumours of soft tissues adjacent to bone which occasionally develop in the same circumstances. Among their cases of radium poisoning, Aub *et al.*[4] reported three examples of epidermoid carcinoma arising in the nasal accessory sinuses or in the temporal bone. Koletsky *et al.*[12] noted squamous cell carcinoma of the nasal region of rats following the administration of phosphorus-32, Kuzma and Zander[18] observed similar squamous cell carcinomas in rats following the administration of strontium-90 (but not in rats receiving calcium-45), and Finkel, Biskis and Scribner[21] have observed similar tumours in mice receiving strontium-90. The present communication directs attention to the occurrence of tumours of the epithelium of the external auditory meatus in certain groups of rabbits receiving strontium-90.

In a study of the effect of a single injection of high dose of strontium-90 (500–1,000 μc./kgm.) in rabbits[13],

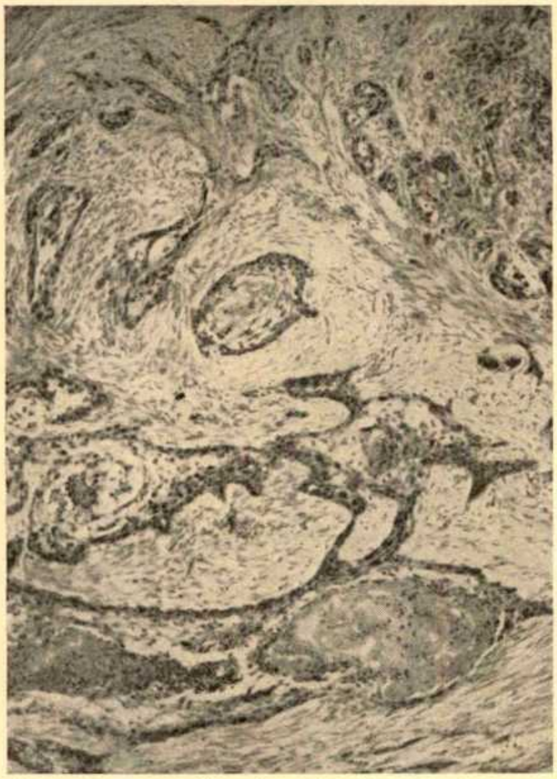

Fig. 1. Squamous cell carcinoma invading soft tissues near temporal bone. (× 66)

it was noted that three of a litter of four rabbits injected with 500 μc./kgm. at the age of 2 days developed symptoms, about 6 months later, attributed to a lesion of the vestibular apparatus. The rabbits were extremely unsteady in gait, they tended to fall over, and carried their heads on one side. Gross examination of the skull and the brain failed to show any obvious abnormalities, except that the external auditory meatus appeared to be blocked with inspissated wax. This proved sterile on culture. The skulls were unfortunately lost before histological examination was made.

A second litter of 4 new-born rabbits, injected with 500 μc./kgm. at 2 days of age, has now been studied. These rabbits all died after a period of 14–22 months: they failed to show evidence of bone damage or tumour formation in the peripheral skeleton, but they presented the same symptoms of ear disease that were noted in the first group. Examination of the external auditory meatus showed that it was occupied by a mass of friable dark wax-like material: histological examination showed the lesions to be squamous carcinomas of the external auditory meatus. In two of the rabbits, invasive squamous cell carcinomas (Fig. 1) of the external auditory meatus were present in each ear: in the remaining two, one ear showed an invasive carcinoma while the other showed a diffuse thickening of the epithelium (Fig. 2) which is regarded as a carcinoma *in situ*. The invasive tumours extended to the soft tissues surrounding the ear, the temporal bone and the cavities of the middle ear.

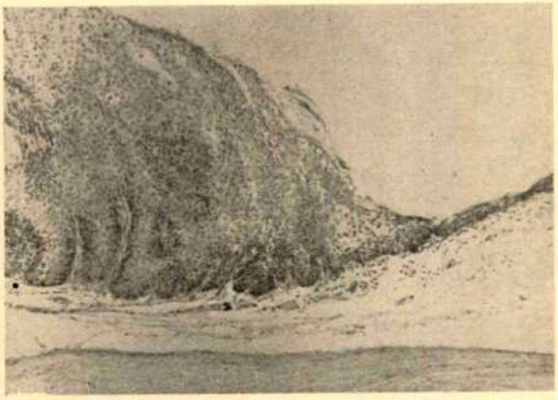

Fig. 2. Thickened (possibly carcinomatous) epithelium in external auditory meatus. (×50)

Table 1

	Dose (μc./kgm. ^{90}Sr)	Survival (months)	Bone tumours	Right ear	Left ear
New-born rabbits					
801*a*	500	7	—	Symptoms of ear disease	
801*c*	500	7	—		
801*d*	500	6	—		
801*b*	500	17	—	No obvious abnormality	
888*a*	500	15	—	Carcinoma	Thickened
888*b*	500	22	—	Carcinoma	Carcinoma
888*c*	500	16	—	Carcinoma	Carcinoma
888*d*	500	15	—	Carcinoma	Thickened
Weanling rabbits					
908	200	19	—	Normal	Normal
909	200	27	—	Carcinoma	Thickened
910	200	24	—	Normal	Carcinoma
802	600	7	++	Thickened	Thickened
902	600	9	++	Carcinoma	Thickened
1-year rabbits					
823	200	19	—	Normal	Normal
870	300	27	—	Normal	Normal
868	400	34	++	Normal	Normal
859	500	24	+	Normal	—
865	500	8	+	Normal	—
807	1,000	7	+	Normal	Normal
847	1,000	6	—	—	Normal
857	1,000	6	+	Normal	—
864	1,000	6	+	Normal	Normal

In order to determine whether epithelial tumours develop in the ears of older rabbits injected with strontium-90, we have examined histological sections from the temporal bones of two other groups of animals (Table 1). The first of these consisted of 5 rabbits injected at the age of 6–8 weeks. Three of these each received 200 μc./kgm. strontium-90 and lived for 19–27 months (908, 909, 910). In two of these animals (one of which had symptoms similar to those present in the previous groups) one ear showed an invasive squamous cell carcinoma of the external auditory meatus, while in the third the epithelium of the external auditory meatus was not abnormal. The two remaining weanling rabbits received 600 μc./kgm. strontium-90 each (802, 902). One, killed at 9 months, had an invasive squamous cell carcinoma of the external auditory meatus on one side and proliferative apparently neoplastic change on the other; the other, killed at 7 months, showed irregular and apparently neoplastic thickening of the epithelium of the external auditory meatus on each side, although no invasive tumour was present.

The second group consisted of 9 rabbits injected at the age of one year with doses of 200–1,000 μc./kgm., and dying 6–34 months later. None of these animals presented symptoms of ear disease, and in none of them were histological abnormalities of the external auditory meatus found.

Spontaneous tumours in this situation do not appear to have been reported in rabbits. It would appear likely that the tumours in the experimental animals resulted from irradiation of the epithelial tissue from strontium-90 deposited in the surrounding and immediately adjacent bone, although the changes we have regarded as carcinoma *in situ* sometimes extend to the epithelium of the outer part of the external auditory meatus at some distance from the temporal bone. In such a situation, the mechanism of tumour induction is not clear: perhaps some migration of the epithelial tissue occurs during growth and development, or the uptake and retention of strontium-90 or its decay product yttrium-90 in other tissues, such as the adjacent cartilage of the pinna, may be concerned.

It is perhaps significant that in the weanling rabbits a number of the ear tumours occurred in animals that had survived longer than others of the same series in which bone tumours had developed, and it may be that the development of malignant change in epithelial tissues adjacent to bone takes longer than in bone itself. In this connexion, it is interesting to note that 2 of the 3 epidermoid carcinomas of nasal accessory sinuses reported by Aub *et al.*[4] in human radium poisoning developed as long as 33 years after exposure to radium, while periods of 15–18 years were concerned with the 6 bone sarcomas reported in the same series.

The retention of strontium-90 and yttrium-90 in the temporal bone and the cartilage of the pinna is being studied further.

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[1] Martland, H. S., *Amer. J. Cancer*, **15**, 2435 (1931).

[2] Looney, W. B., and Woodruff, L. A., *Arch. Path.*, **56**, 1 (1953).

[3] Looney, W. B., Hasterlik, R. J., Brues, A. M., and Skirmont, E., *Amer. J. Roentgenol.*, **73**, 1006 (1955).

[4] Aub, J. C., Evans, R. D., Hempelmann, L. H., and Martland, H. S., *Medicine*, **31**, 221 (1952).

[5] Sabin, F. R., Doan, C. A., and Forkner, C. E., *J. Exp. Med.*, **56**, 267 (1932).

[6] Schürch, O., and Uelinger, E., *Arch. klin. Chir.*, **183**, 704 (1935).

[7] Hellner, H., *Arch. klin. Chir.*, **189**, 706 (1937).

[8] Dunlap, C. E., Aub, J. C., Evans, R. D., and Harris, R. S., *Amer. J. Path.*, **20**, 1 (1944).

[9] Petrov, N. N., Krotkina, N. A., Vadova, A. V., and Postnikova, Z. A., "Dynamics of the Origin and Development of Malignant Growths in Experiments on Monkeys" (Russian) (U.S.S.R. Acad. Med. Sci. Press, Moscow, 1952).

[10] Heller, M., "Histopathology of Irradiation", Ed. Bloom, W. (McGraw-Hill, New York, 1946).

[11] Brues, A. M., Lisco, H., and Finkel, M., "Carcinogenic Action of Some Substances which may be a Problem in Certain Future Industries", Rep., U.S. Atomic Energy Commission (1946).

[12] Koletsky, S., Bonte, F. J., and Friedell, H. L., *Cancer Res.*, **10**, 129 (1950).

[13] Owen, M., Sissons, H. A., and Vaughan, J., *Brit. J. Cancer*, **11**, 229 (1957).

[14] Downie, E. D., Macpherson, S., Ramsden, E. N., Sissons, H. A., and Vaughan, J., *Brit. J. Cancer*, **13**, 408 (1959).

[15] Anderson, W. A. D., Zander, G. E., and Kuzma, J. F., *Arch. Path.*, **62**, 262 (1956).

[16] Cherkasskii, L. A., *Vop. Onkol.* (Russian), **2**, 275 (1956).

[17] Litvinov, N. N., *Bjull. ekspn. Biol. Med.* (Russian), **8**, 62 (1956); *Vop. Onkol.* (Russian), **2**, 285 (1956).

[18] Kuzma, J. F., and Zander, G. E., *Arch. Path.*, **63**, 198 (1957).

[19] Skoryna, S. C., and Kahn, D. S., *Cancer*, **12**, 306 (1959).

[20] Holgate, W., *J. Dent. Surg.* (in the press).

[21] Finkel, M. P., Biskis, B. O., and Scribner, C. M., Proc. 2nd Intern. Conf. Peaceful Uses Atomic Energy, Geneva (1958).

Incorporation of Tritiated Uridine into Amphibian Eggs

THE incorporation of labelled precursors into amphibian embryos is limited by the presence of a coat which covers the embryo and which renders the embryo impermeable to a large number of substances. One of them is uridine. However, we found that embryonic cells the coat of which has been disintegrated by placing them in a medium totally devoided of calcium incorporate tritiated uridine very rapidly, while the control embryos do not. Using this procedure, we have been able to follow the incorporation of this substance into dissociated embryonic cells at different stages of development.

Eggs removed at the desired stages were decapsulated. These eggs, still in their vitelline membrane, were cultivated in the following medium: sodium chloride (17 per cent), 20 ml.; potassium chloride (0·5 per cent), 10 ml.; *tris*, 560 mgm.; water, 966 ml.; *N* hydrochloric acid, 4 ml.; citrate, 5 gm.

In this medium, the embryos disaggregate into a mass of dissociated cells surrounded by the vitelline membrane.

After dissociation, the cells were replaced in the same medium without calcium, but deprived of citrate. The incorporation of uridine was followed by the autoradiographic technique of A. Ficq[1], after fixation by freeze-substitution.

At the blastula and young gastrula stages, the incorporation of tritiated uridine is exclusively nuclear. Treatment of the sections with deoxyribonuclease before application of the photographic emulsion completely removes the radioactivity: on the other hand, treatment of the sections with water or ribonuclease has no effect on the radioactivity. Therefore, uridine seems to be incorporated into deoxyribonucleic acid. This incorporation probably occurs at the level of the pyrimidine bases of deoxyribonucleic acid, because hydrolysis of the sections with *N* hydrochloric acid for 5 min. at 60° has no effect on the radioactivity. During gastrulation and neurulation, nuclear activity remains intense. A greater heterogeneity in the distribution of radioactivity among the embryonic nuclei is observed. At the same time, radioactivity becomes conspicuous in the cytoplasm. Cytochemical tests indicate a change in the nature of the substances incorporating uridine. The nuclei, in slides treated with ribonuclease or *N* hydrochloric acid at 60°, now lose about half their radioactivity. After incubation of the sections in the presence of deoxyribonuclease, 30–40 per cent of the radioactivity remains in the nuclei. It should be mentioned that all the cells do not behave in exactly the same manner when they are submitted to this treatment. At the onset of segmentation of the egg, the importance of the coat is still such that penetration of the precursor is impossible even in the presence of citrate. Unsegmented eggs incubated in the presence of uridine and citrate for about 20 hr. exhibit nuclear incorporation in the daughter cells. At this stage, the localization of the tracks is either nuclear or chromosomal.

In summary, until the neurula stage, nuclear incorporation predominates over cytoplasmic incorporation. Uridine incorporation occurs principally into deoxyribonucleic acid during segmentation. After gastrulation, there is an incorporation of uridine into ribonucleic acid (both nuclear and cytoplasmic) as well.

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[1] Ficq, A., *Arch. Biol.*, **66**, 508 (1955).

BIOLOGY

Birefringence of Enamel Organic Matrix

To date little attention has been given to the appearances of the enamel organic matrix when examined between crossed nicols, and the few published observations show considerable disagreement. Cape and Kitchin[1], and Poole[2] reported that the enamel organic matrix is without intrinsic birefringence. Keil[3] and Schmidt[4] reported a weak positive intrinsic birefringence—the sign being described with respect to the long axis of the enamel prisms. Rushton[5] stated that this positive birefringence is strong, in fact stronger than the negative intrinsic birefringence of the mineral matter present in the mature tissue.

During current studies on the optical properties of dog enamel, results were obtained which support the views of Keil and Schmidt. In these studies ground sections, approximately 300µ in thickness, prepared from the developing maxillary permanent incisors of a two months old dog were demineralized in a formic and citric acid mixture. The demineralized sections were first imbibed for periods of 24 hr. in each fluid of a series prepared by mixing varying proportions of cedarwood oil ($n_D = 1{\cdot}505$) and monobromnaphthalene ($n_D = 1{\cdot}659$), and then examined between crossed nicols in monochromatic light—a sodium lamp being used as light source. The refractive indices of the imbibition fluids, determined with an Abbe refractometer, ranged from 1·505 to 1·598 by intervals of approximately 0·01. The retardation at selected points on the specimens was measured in each fluid by means of a Berek compensator.

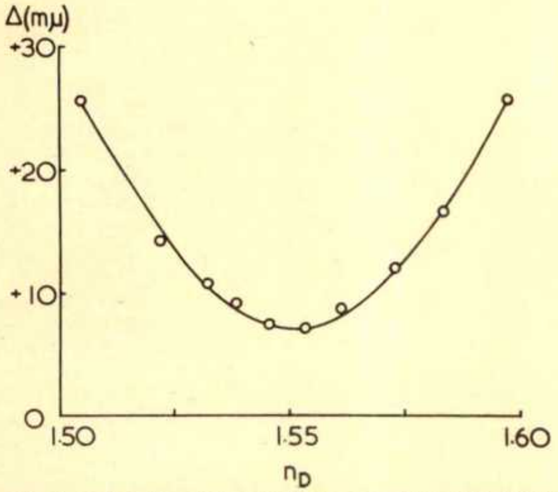

Fig. 1. Retardation of the enamel organic matrix as a function of the refractive index of the imbibition fluid

The type of imbibition curves thus obtained (Fig. 1) clearly indicates that in demineralized developing dog enamel organic matrix there is an interaction between positive form birefringence and positive intrinsic birefringence. Allowing for possible variations in prism direction giving rise to a crossed lamellar system[6], the strength of the positive intrinsic birefringence appears to be not less than 0·00002. Further, the refractive index of the organic matrix at this stage of development is shown by the imbibition curves to be about 1·55—a value confirmed by Becke line tests.

Thanks are due to Prof. L. R. Moore for making available the facilities to conduct this study in the Geology Department.

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[1] Cape, A. T., and Kitchin, P. C., *J. Amer. Dent. Ass.*, **17**, 193 (1930).

[2] Poole, D. F. G., *Quart J. Micro. Sci.*, **98**, 349 (1957).

[3] Keil, A., *Z. Zellforsch.*, **22**, 633 (1935).

[4] Schmidt, W. J., *Z. Zellforsch.*, **49**, 319 (1959).

[5] Rushton, M. A., *Brit. Dent. J.*, **67**, 1 (1939).

[6] Frey-Wyssling, A., "Submicroscopic Morphology of Protoplasm and its Derivatives" (Elsevier, Amsterdam, 1948).

Placental Grafts in Rats

DURING studies of experimental infection of the rat placenta attempts were made to transplant placental tissue from the uterus to the mother's omentum. Grafts of the allanto-chorion were not successful; but the yolk sac membrane, when transplanted at 15 days pregnancy, grew in a most remarkable manner.

After a number of pilot experiments the following technique was adopted. The abdominal cavities of rats (Albino Wistar strain) at 15 days pregnancy were opened under open ether anæsthesia. The uterus with its contained conceptuses was removed, but the ovaries left intact. A portion of yolk sac was carefully dissected from a placenta and returned to the mother's peritoneal cavity where it was wrapped in the omentum. The abdominal wound was then closed.

Of 12 rats treated in this manner all except one showed evidence of transplant growth at autopsy 4, 8 or 12 weeks later. Seven transplants grew into large mucus-containing cysts, the largest being more than 2 in. in diameter (Fig. 1). On histological examination a variety of tissues were frequently found together in the same graft. These included epidermoid cysts containing keratin and hair, hair follicles, sebaceous and sweat glands, mucus-secreting epithelium, whorled masses of mesenchyme connective tissue, smooth and striated muscle, cartilage and bone. The mucus-secreting epithelium was usually organized with other tissues so as to resemble gut wall, in that there were villi and surrounding layers of smooth muscle (Fig. 2).

The possibility of dependence of these grafts on hormones has been briefly investigated; removal of the

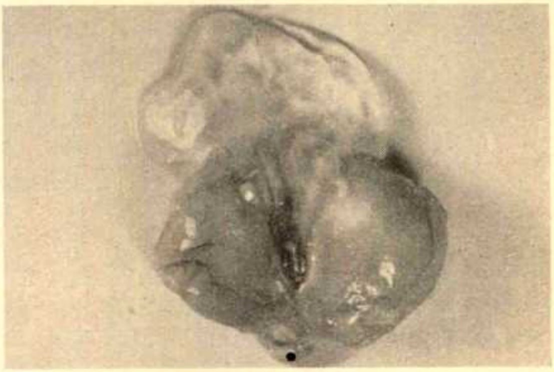

Fig. 1. Rat yolk sac graft to omentum, 12 weeks after transplantation. It has grown to more than 2 in. in diameter and contains large quantities of mucus

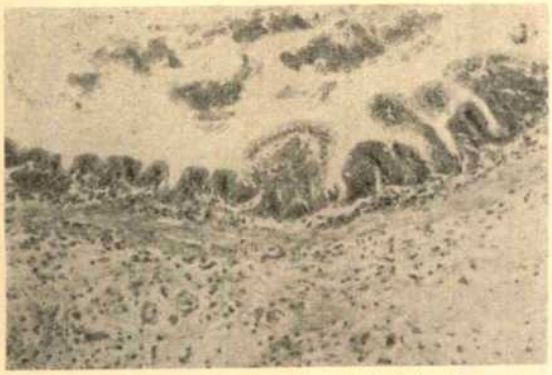

Fig. 2. As for Fig. 1. The wall of the graft is lined by mucus-secreting epithelium beneath which is connective tissue and layers of smooth muscle cells. Hæmatoxylin and eosin. (×150)

ovaries did not prevent successful transplantation, but the grafts did not grow to the same extent. Similar results occurred with homografts into 4 non-pregnant intact females. Homografts to 4 males grew in only 2 animals, where they produced just small quantities of mesenchyme connective tissue and tiny epidermoid cysts.

The variety of tissues produced in these grafts suggests that after transplantation to an unfamiliar site the endoderm and mesoderm cells of the rat yolk sac are capable of diverse differentiation.

As we have not found any previous account of similar transplants of placental tissue in rats we wish to place these initial results on record. Details and further experiments now in progress will be reported elsewhere.

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In vitro Culture of *Musa balbisiana* Colla Embryos

THE culture of excised embryos has been reported for many plants[1], but, to our knowledge, there are no reports concerning the successful cultivation of embryos of seeded bananas. In the course of investigations on the rhizosphere of bananas, it became necessary to obtain plants of *Musa balbisiana* Colla, the roots of which were free of micro-organisms. The germination of intact seeds of this variety under sterile conditions was generally unsuccessful; hence, the possibility of obtaining sterile plants from excised embryos was investigated.

Preliminary studies were conducted with a modified Knudson's[2] medium, containing 2 per cent sucrose and 1·2 per cent agar. Intact seeds were surface sterilized in filtered $CaOCl_2$ (10 gm. $CaOCl_2$ in 150 ml. of distilled water)[2] *in vacuo* for 45 min. The embryos were removed aseptically and placed on 30 ml. of medium in 4 oz. wide-mouth screw-cap jars. Enlargement of the embryos was followed by leaf and root development. The roots, however, failed to penetrate the medium, and growth of the plantlets ceased after approximately 6 weeks, resulting ultimately in death of the plants. When young plantlets were transferred from the Knudson medium to that of Randolph and Cox[3], which contained only 0·7 per cent agar, root development improved and the roots penetrated into the medium.

In all current studies the Randolph and Cox medium is being used. The agar content of the medium has been further reduced to 0·5 per cent, since more rapid growth and healthier plants have been obtained with this concentration. Sucrose was found to be better than glucose or fructose as a carbohydrate source and 4 per cent was the optimum level. Intact seeds are surface sterilized by agitating for 30–35 min. in $CaOCl_2$ on a wrist-action shaker; the use of vacuum was found to be unnecessary in order to obtain sterility. The seeds are split longitudinally and the embryos aseptically removed. Excised embryos may be immersed in $CaOCl_2$ for up to 5 min. without deleterious effects, in agreement with Tukey's[4] experience with the embryos of stone fruits. In early experiments, the embryo, after enlargement, was re-oriented on the agar so that the cotyledon was placed in contact with the medium to provide a larger surface area for absorption. Repositioning was unnecessary when the low agar medium was used, since the weight of the embryo embedded the cotyledon into the soft agar.

Callus-like growth generally is evident 5–6 days after excision, followed by the appearance of the coleoptile and roots. Often, delicate hair-like growths arise from the surface of germinating embryos prior to the appearance of the coleoptile and roots. Two weeks after excision, the first leaf and roots have developed. A six-week old plantlet is illustrated in Fig. 1.

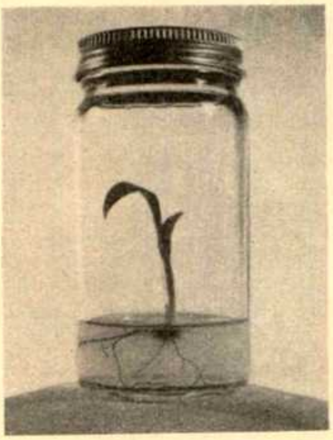

Fig. 1. *Musa balbisiana* Colla. Six-week-old plantlet developed in embryo culture

Plants developed from mature embryos, excised from seeds which were stored for periods of 3 weeks to 1½ years after harvest, were grown aseptically in the original culture vessels for 3 months. They were then transplanted to growth tubes wherein the roots have been maintained under sterile conditions for an additional 3 months. These plants are approximately 8 in. tall and have 8–10 fully developed leaves and well-developed root systems. The plants are smaller than seedlings of the same age which developed in soil under greenhouse conditions from intact seeds, but otherwise they appear to be similar in all respects.

The application of embryo culture to studies in plant breeding and nutrition have been pointed out by Crocker and Barton[1]. Our experiments have demonstrated that a wild relative of the commercial banana can be grown successfully from mature embryos and that the technique could possibly be applied to other varieties of bananas.

Studies are in progress on the nutrition, physiology, and morphology of *M. balbisiana* plants derived from embryos, results of which will be presented elsewhere.

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[1] Crocker, W., and Barton, Lela V., "Physiology of Seeds" (Chronica Botanica Co., Waltham, Mass., 1953).

[2] Knudson, L., *Amer. J. Bot.*, **37**, 241 (1950).

[3] Randolph, L. F., and Cox, L. G., *Proc. Amer. Soc. Hort. Sci.*, **43**, 283 (1943).

[4] Tukey, H. B., *Bot. Gaz.*, **99**, 630 (1938).

SOIL SCIENCE

Weathering of Fly Ash

A CONSIDERABLE body of information is now available regarding the minerals present in fresh fly ash which are available to plants[1,2]. During combustion of the coal, adventitious clay minerals such as kaolinite are broken down, and a material is produced containing all the mineral constituents of coal, but with a very low cation exchange capacity (3 m.equiv. per 100 gm.). Many metal oxides are embedded in the glassy phase of the fly ash and may be released slowly over a period of time. In considering the possibilities of establishing crops or plant cover on fly ash deposits, it seemed worth while, therefore, to obtain information on the rate of change of selected constituents. Of particular interest were the elements, for example, boron and aluminium, that are present in quantities likely to be injurious to plants. In the studies to be briefly described here determinations were also made of phosphorus, potassium, calcium and iron in order to obtain any information regarding any likely change in fertilizer requirements as the age of the ash increases.

Ash tip areas covering an age-range of 0–25 years were available at Hams Hall power station and analyses have been made of samples collected from them. Phosphorus, iron and aluminium were determined in extracts obtained using Morgan's reagent. Boron, potassium and calcium were determined in water extracts using standard time procedure as for water-soluble soil constituents. These results, plotted against the age of the ash tip, are shown in Figs. 1 and 2 and indicate a rapid initial decrease in available concentration.

In order to see how far the changes could be due simply to leaching, columns 5 cm. in diameter and containing 25 gm. of fresh ash were set up and leached under a constant head of about 1–5 cm. of water and at a rate of about 1 ml./min. The leachate was collected in aliquots of 40 ml. for analysis. The curves obtained for potassium, calcium and boron (Fig. 3) are in agreement with field observations: a rapid initial decrease followed by a steady release of the elements. No phosphate, aluminium or iron was detected in the leachate. It was also noted that in the weathering of field samples the decrease in available iron was molecularly equivalent to the fall in available phosphate and that the fall in available calcium was similarly equivalent to the fall in available aluminium. These facts and the laboratory leaching curves suggest that, for these elements, the decrease in available concentration might be due to chemical fixation rather than to leaching out.

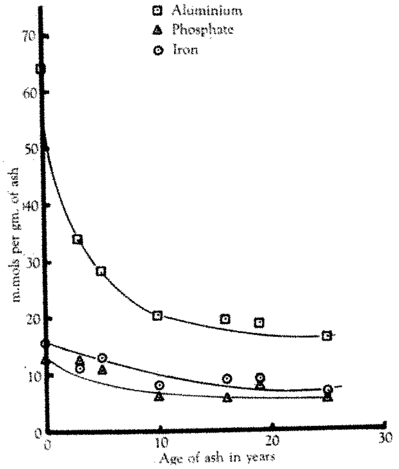

Fig. 1. **Amounts of aluminium, iron and phosphate in Morgan's extract of P.F. ash of varying age**

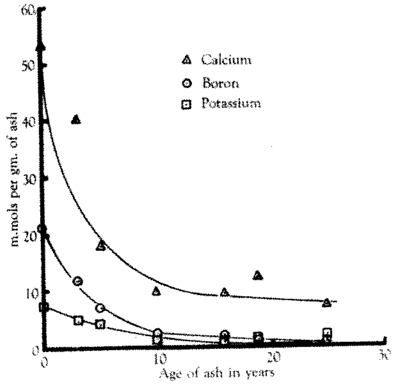

Fig. 2. **Amounts of calcium, boron and potassium in water extract of P.F. ash of varying age**

The equivalence of the change in available calcium and aluminium would suggest the formation of a dicalcium aluminate but there is as yet no evidence for this. X-ray studies[3] indicate the formation of a tetra calcium aluminate and some calcium sulphoaluminate after prolonged hydration and it seems likely that some interaction between calcium and aluminium takes place. The rapid loss of soluble calcium found in the leaching experiments would not be likely to occur to the same extent under field conditions as the relatively soluble calcium hydroxide would be rapidly carbonated and fixed in an insoluble form.

The fall in available concentration of elements injurious to plant growth on the fly ash, namely boron and aluminium, is also of particular interest. It may be seen from Figs. 1 and 2 that these elements fall rapidly from a high injurious level to a level approaching that of a normal soil (for example, 1 p.p.m. water-soluble boron[4]). Fig. 4 shows how the boron content of wild white clover grown on ash falls with increasing age of the ash used. It may be concluded that although the range of possible crops

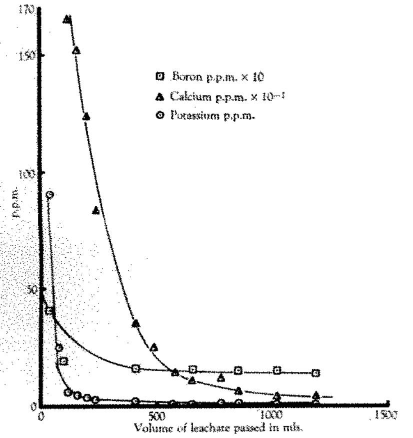

Fig. 3. Laboratory leaching curves

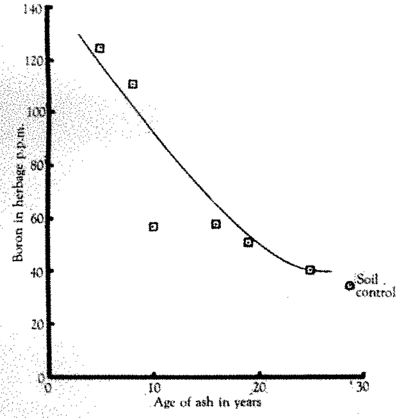

Fig. 4. Boron in plant tissue grown upon ash of varying age

is limited during the first few years of the life of a fly ash tip a considerably wider range of crops may be used as the ash weathers.

In using already existing ash tips of different ages for the investigation of weathering it has been assumed that (*a*) the coal source and composition had remained reasonably constant over the 25-year period and (*b*) that improved methods of coal grinding, resulting in the production of a finer ash, have not significantly altered the availability of elements as for example by increasing surface area. The latter possibility cannot be ignored but the agreement between the column leaching experiments and the field observations indicates that a true picture of weathering under field conditions has been obtained. The samples examined were procured from areas with free drainage and no evidence of waterlogging. The rate of decrease in soluble constituents in areas with a high water table and/or impeded drainage may well be considerably lower.

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[1] Rees, W. J., and Sidrak, G. H., *Plant and Soil*, **8**, 141 (1956).

[2] Holliday, R., *et al.*, *Nature*, **181**, 1079 (1958).

[3] Private communication from Central Electricity Generating Board Research Laboratories, Leatherhead.

[4] Bear, F. E. (Ed.), "Chemistry of the Soil", 329 (Reinhold Publishing Co., New York, 1955).

PATHOLOGY

Periosteal Osteoclasts, Skeletal Development and Ageing

As far back as 1873, Kölliker[1] observed the existence of osteoclasts in the periosteum of normal bone. Lacroix[2] also made several references to the presence of periosteal osteoclasts during development of the perichondral collar of long bones. Quantitative information, however, is not available concerning the size of the population of periosteal osteoclasts and the appearance, disappearance, and stability of the population during ageing. The importance of the presence of these cells at periosteal sites has been largely neglected. Environmental conditions which are found in the periosteum make for an excellent study of osteoclastic origin, function, fate and biochemical nature.

Jackson[3] and Jordan[4] have reported mitosis in the nuclei of osteoclasts. Amitosis has also been noted by Kolliker[5]; however, these events are extremely rare, and cannot account for the rapid appearance and disappearance of large numbers of osteoclasts. Kölliker maintained that osteoclasts originated from the fusion of osteoblasts. More recently Heller *et al.*[6] described the formation of osteoclasts from osteoblasts or osteocytes under the influence of toxic doses of parathormone.

Because of the nature of the periosteum, it was felt that a study of periosteal osteoclasts may reveal information concerning the origin and fate of osteoclasts under normal conditions, which could not be readily observed in the more complex environment of the metaphysis. For a better comprehension of bone development and ageing, population studies were made from the left femora of 45 female mice of the Brookhaven National Laboratory strain of Swiss Albino. The right femur of each mouse was fractured by digital pressure applied to the mid-shaft under ether anæsthesia. The femora were fractured at 1, 5, 8, 26, and 52 weeks of age, and the mice were killed 24 hr., 1 week, and 2 weeks post-fracture. One hour before they were killed, they received a subcutaneous injection of tritium-labelled thymidine in sterile aqueous solution (0·5 μc./gm. body-weight). Stripping film autoradiographs were prepared from paraffin sections of whole decalcified femora. Additional sections were stained routinely with hæmatoxylin and eosin.

The tritium-labelled thymidine technique and interpretation of data are based on the successful studies employed by Hughes *et al.*[7], Cronkite *et al.*[8,9], Bond *et al.*[10], and Quastler and Sherman[11].

The total population of periosteal osteoclasts was found to be much larger in animals the femora of which were undergoing active bone development and remodelling than in older mice. Counts made of the population of periosteal osteoclasts of young mice revealed extensive fluctuations within very short periods of time (Table 1 and Fig. 1). A more stable population was exhibited by older animals, the total osteoclastic population of which was much lower than the younger mice. Considerable differences were found between counts made from the anterior and posterior aspects of the femur. A shift in the position of the major population of osteoclasts was observed at least 3 times during the ages studied. This shift is indicated in Fig. 1 at the points of intersection.

It is interesting to point out that on numerous occasions in animals of different ages cells were

Table 1. AVERAGE NUMBER OF OSTEOCLASTS PER UNIT DISTANCE* OF PERIOSTEUM COVERED AT THE DISTAL EPIPHYSEAL ASPECT OF MOUSE FEMUR

Age (weeks)	1	2	3	5	6	7	8	9	10	26	27	28	52	53	54
Anterior + posterior counts	0·08	0·35	0·51	0·69	0·26	0·98	0·46	0·30	0·34	0·18	0·07	0·10	0·02	0·02	0·02
Anterior counts	0·13	0·54	0·61	0·55	0·12	0·67	0·24	0·11	0·47	0·06	0·01	0·10	0·01	0·01	0·01
Posterior counts	0·04	0·29	0·41	0·79	0·40	1·30	0·70	0·47	0·21	0·30	0·13	0·10	0·02	0·04	0·03

* An average of 173 unit distances covered per age group.

observed having a size comparable to osteoblasts. Except for the presence of a single nucleus, the cells answered to the description of osteoclasts in all cytological respects. The mononucleated state was verified from serial sections. These mononucleated cells, which in many instances resided in well-formed Howship's lacunæ, exhibited an eosinophilic, foamy cytoplasm, lacking in the presence of a juxtanuclear vacuole. A juxtanuclear vacuole is seen in active osteoblasts. Cells of this description were never encountered in the metaphyses. Ch'uan[12] has observed mononucleated osteoclasts in the jaws of a two-day-old hamster and the stripped periosteum of a newly born goat.

Several foci of osteoblasts arranged in tight clusters were observed at ages where active periosteal osteoclast formation was taking place. Individual cells making up these clusters finally lost the identity of their cytoplasmic borders, cytoplasmic basophilia, and juxtanuclear vacuoles. The usual population of periosteal osteoblasts was reduced in the immediate areas where osteoclasts were being formed.

Autoradiographs of intact femora revealed an essentially weakly labelled periosteum. The highest labelling occurred at the perichondral region. Osteogenic cells of the osteogenic layer of the periosteum near the fibrogenic layer were labelled most frequently. Osteoblasts, however, were rarely seen labelled. Osteoclasts, on the other hand, were never seen labelled above background (Fig. 2). This is also true of metaphyseal osteoclasts. With increasing age labelling of periosteal cells became more infrequent.

Within 24 hr. after fracture and at all ages studied, the periosteum became increasingly labelled about the fracture site. In older animals, however, the overall labelling was not as high as the level found in younger mice. Periosteal labelling continued to increase until the osteogenic cells of the fracture callus differentiated into cartilage cells (Fig. 3). Diminished periosteal labelling was noted when the active replacement of the cartilagenous callus by endochondral ossification took place. With increasing age there was a retardation of fracture healing in mice. Reduction in the rate of bone repair has been observed and reported for rat femora[13,14]. The degree of labelling was dependent upon the stage of repair that the fracture was in, and the age of the animal.

No uptake of tritium-labelled thymidine was observed in the numerous osteoclasts which appeared during fracture healing. Labelling of osteocytes was never seen.

The results showed that during the remodelling of the ends of the femur, the population of osteoclasts present in the periosteum fluctuated radically within short periods of time. Since active formation of periosteal osteoclasts was observed from the fusion of osteoblasts, and degenerating osteoclasts were

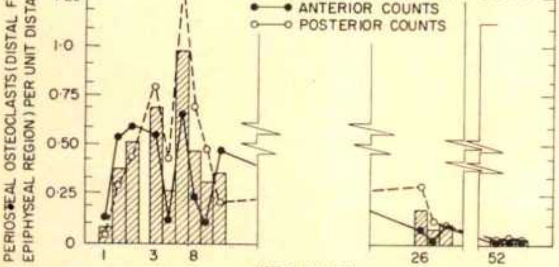
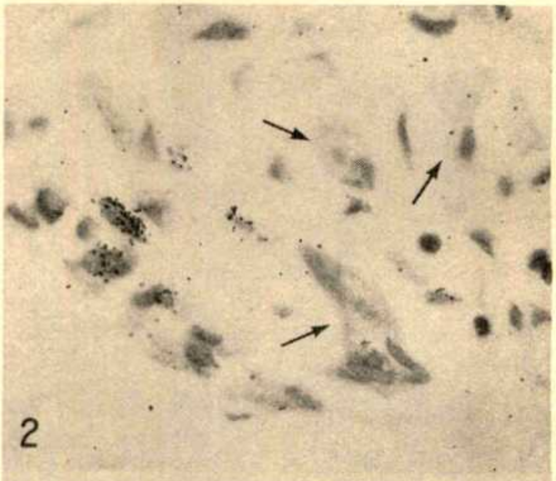


Fig. 1. Graph showing a comparison between the size of the osteoclastic population of the anterior and posterior aspects of the periosteum in the distal end of the femur at different ages



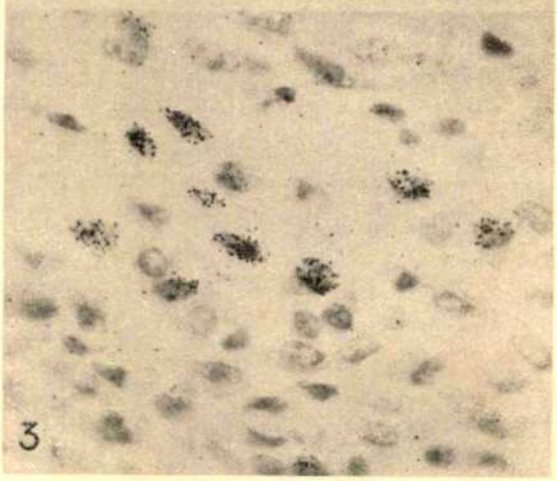


Fig. 2. Photomicrograph revealing tritium-labelled thymidine marking of osteoblasts during active fracture healing in a 2-week-old-mouse the femur of which had been fractured 1 week prior. Arrows point to non-labelled osteoclasts (× c. 534 oil)

Fig. 3. Photomicrograph revealing tritium-labelled thymidine marking of osteogenic cells of the periosteum active in the production of the fracture callus. The cells are undergoing transformation into chondrogenic cells in a 2-week-old mouse the femur of which had been fractured 1 week prior (× c. 560 oil)

not readily found, a reduction in population within one week's time is believed to have occurred as a result of the dissociation of osteoclasts into precursor cells. It is possible that the population of osteoclasts migrated from one position to another. Had this migration taken place, there would have been fewer periosteal cells present in such regions, unless the other periosteal cells filled the gap. No deficiency was encountered in the population of periosteal osteoblasts or osteogenic cells at that time. At a point, however, when the total (anterior and posterior) population of osteoclasts diminished rapidly, the presence of degenerating cells should have been seen. Nevertheless, degenerating osteoclasts were not encountered. Autoradiographic results indicated that there was no uptake of tritium-labelled thymidine by periosteal osteoclasts and hence there occurred neither deoxyribonucleic acid synthesis nor reproduction by mitotic division.

The observations indicate that periosteal osteoclasts derive their origin from osteoblasts and their fate is to dissociate into precursor cells. It appears that with increasing age beyond the period of active bone remodelling, there is a decrease in the population of osteoclasts; however, changes in age in individual osteoclasts were not seen.

I wish to acknowledge the suggestions of Dr. E. P. Cronkite, and the technical assistance of Miss M. Pavelec.

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[1] Kölliker, A., "Die normale Resorption des Knochengewebes und ihre Bedeutung für die Entsehung der typischen Knochenformen" (Vogel, Leipzig, 1873).

[2] Lacroix, P., "The Organization of Bones" (The Blakiston Co., Philadelphia, 1951).

[3] Jackson, C. M., *Archiv Anatomie u. Entwicklungsgeschichte*, **33**, 70 (1904).

[4] Jordan, H. E., *Anat. Rec.*, **30**, 107 (1925).

[5] Kölliker, A., "Handbuch der Gewebelehre des Menschen", 6th Ed., 1 (W. Engelman, Leipzig, 1889).

[6] Heller, M., McLean, F. C., and Bloom, W., *Amer. J. Anat.*, **87**, 315 (1950).

[7] Hughes, W. L., Bond, V. P., Brecher, G., Cronkite, E. P., Painter, R. B., Quastler, H., and Sherman, F. G., *Proc. U.S. Nat. Acad. Sci.*, **44**, 476 (1958).

[8] Cronkite, E. P., Fliedner, T. M., Bond, V. P., Rubini, J. R., Brecher, G., and Quastler, H., *Ann. N.Y. Acad. Sci.*, **77**, 803 (1959).

[9] Cronkite, E. P., Fliedner, T. M., Bond, V. P., and Robertson, J. S., in "The Kinetics of Cellular Proliferation", F. Stohlman, editor (Grune and Stratton, Inc., New York, 1959).

[10] Bond, V. P., Fliedner, T. M., Cronkite, E. P., Rubini, J. R., and Robertson, J. S., *ibid.*

[11] Quastler, H., and Sherman, F. G., *Exp. Cell Res.*, **17**, 420 (1959).

[12] Ch'uan, C. H., *Anat. Rec.*, **49**, 397 (1931).

[13] Tonna, E. A., *Nature*, **181**, 486 (1958).

[14] Tonna, E. A., *J. Gerontol.*, **14**, 159 (1959).

ARCHÆOLOGY

An Alternative Interpretation of the Supposed Giant Deciduous Hominid Tooth from Olduvai

DR. L. S. B. LEAKEY has reported[1,2] the discovery at Olduvai of what he regards as a large second lower deciduous molar of a hominid exhibiting affinities with *Sinanthropus* and the Heidelberg jaw, but not with australopithecines. Associated with it was a worn deciduous canine. The purpose of this communication is to show that the molar is, in fact, a permanent upper tooth, probably of an australopithecine.

The molar appears to be a somewhat aberrant specimen, since the morphologically less-stable areas —anterior and posterior foveæ and either the lingual or buccal faces, depending on whether the tooth is maxillary or mandibular—have many accessory cusps.

Assuming the tooth to be mandibular, as Leakey does, and ignoring the accessory cusps for the present, it is not possible to fit the main cusp or fissure patterns into either of the two common types for hominoid mandibular molars: the five- or four-cusped types. The relationship of the cusps to each other is wrong: the buccal cusps are too far back relative to the lingual ones. Furthermore—a major difficulty in interpreting the tooth as mandibular—the anterior buccal cusp ('protoconid') and the distal lingual cusp ('entoconid') are linked by an almost cusp-high crest. This divides the fissure system into two quite separate parts, while even in very worn mandibular molars of earlier hominids the fissure system separates these two cusps deeply and is not divided into two portions. Also, this oblique, high crest with a deep fissure posterior to it cutting deeply between the 'protoconid' and 'hypoconid', distinctly sets off an almost symmetrical anterior triangle of major cusps from a single, semi-isolated major cusp posteriorly. This is a feature not found on mandibular molars.

On the other hand, the high, oblique crest in this position on the crown, the anterior trigon of three major cusps and a postero-medial semi-isolated cusp (hypocone) are not merely common on maxillary molars but also are regarded as being the standard pattern for them. Leakey regards two large accessory cusps in the posterior fovea as very unlikely on a maxillary tooth. Fig. 1 (*C*) illustrates an almost identical condition in an upper molar; one *Australopithecus* upper molar from Sterkfontein has no less than five accessory cusps in this position.

The crown morphology is thus clearly in favour of this tooth being maxillary. The disposition of the roots provides an absolute check, since early hominid deciduous or permanent mandibular molars have two roots, an anterior and a posterior, while maxillary ones have a single lingual and two buccal roots. There can thus be no confusion. Although damaged, the Olduvai specimen clearly has two buccal roots and a larger, single lingual one. For comparison a maxillary permanent australopithecine molar in a similar state of preservation is shown in Fig. 1 (*D*) along with the entirely different condition in an australopithecine deciduous lower second molar (Fig. 1, *F*). The roots of the Olduvai specimen manifestly could not belong to a mandibular tooth.

Leakey regards the tooth as deciduous because of: (i) divergent roots, (ii) constriction of the neck, (iii) nature of the roots, and (iv) association with a lower deciduous canine.

Divergent roots and a constricted neck, in earlier hominid molars, do not necessarily indicate deciduous teeth, since the permanent teeth commonly have both features well developed. This is well shown by the extensive collections of australopithecine teeth now known. As already indicated in the case of (iii), the nature of the roots is more consistent with the specimen being a permanent upper tooth. Finally, (iv) is a broken reed. If this tooth is regarded as a second lower deciduous molar, then it is highly

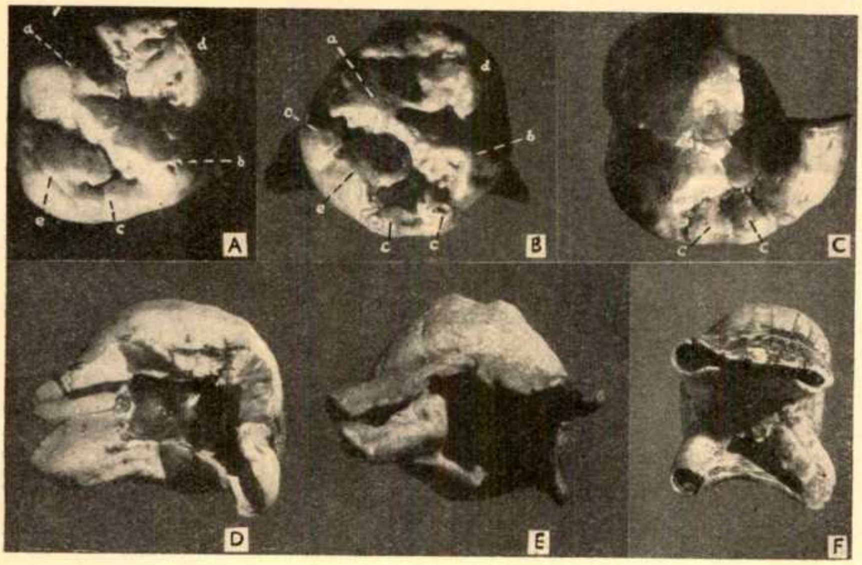

Fig. 1. (*A*) and (*C*) Occlusal views of australopithecine maxillary molars. (*B*) Occlusal view of Olduvai molar. View of the root system (damaged) of an australopithecine maxillary molar (*D*) and of the Olduvai molar (*E*). (*F*) View of root system of an australopithecine deciduous mandibular molar. In all cases the lingual side is to the left. In the upper row the anterior end is at the top and in the lower row at the bottom. In (*A*) and (*B*) the high oblique crest links *a* and *b*; the trigon is formed by *a*, *b* and *d*. *a*, Protocone; *b*, metacone; *c*, accessory cusps; *d*, paracone; *e*, hypocone

unlikely that it belonged to the same individual as the deciduous canine. The latter is well worn, and had already lost about half the crown height. At this stage, dm_2 of the individual would already have been appreciably worn, not virtually unworn as is the Olduvai molar. Furthermore, at that stage permanent molars with fully formed crowns would also have been present. On the other hand, while the deciduous canine is only very slightly larger than the observed range of the very small collection of australopithecine equivalents, the molar is appreciably larger than the largest australopithecine dm_2 known, but is well within the observed range for permanent molars.

I believe, therefore, that the Olduvai molar is neither deciduous nor mandibular, but is a permanent upper tooth, probably australopithecine.

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[1] Leakey, L. S. B., *Nature*, **181**, 1099 (1958).

[2] Leakey, L. S. B., *Illus. Lond. News*, **232**, 1104 (1958).

IN my original paper I was at pains to point out that of a number of dental surgeons and anatomists to whom I had then showed this tooth, no two were agreed upon what its place was in the dental series. Since that time I have had many other views put to me in letters and also verbally. Clearly, the tooth is so aberrant there is little hope of agreement.

It seems to me to be immaterial, at the moment, as to which opinion is correct. What is certain and important is that it is: (*a*) a very large *hominid* molar; (*b*) that it was found within a few inches of a small deciduous canine tooth and within a few minutes; (*c*) that it comes from the base of Bed II, Olduvai, associated with Stage I of the Chellean culture, and was found on a living floor; (*d*) that the relative size of the molar to the canine is comparable to that seen in *Zinjanthropus*, and to a lesser extent in *Paranthropus* and *Australopithecus*.

I still retain my opinion that, in spite of its unusual form, it is a lower deciduous molar, and the fact that I have now three modern hominid lower deciduous molars (taken from mandibles) that similarly have three roots and not two, and set in just this manner, shows that Robinson's reliance on the number of roots, to clinch his argument, is not realistic. Similarly, while I agree (and also stressed in my original paper) that if the tooth is, as I believe, a lower molar, it does *not* show either of the two common types of cusp patterns, the cusp pattern which it does exhibit is, nevertheless, one which does occur in Hominids, both recent and extinct, as I showed before.

Since that article was written, I have found that the same pattern occurs in many other modern mandibular molars, and we must indeed now regard it as a third fairly common pattern, since I found it in 15 per cent of a random sample of mandibles.

Among the many anatomists who have now seen the fossil tooth in question, there are some who agree wholly with me; there are others who agree with Robinson, and there are yet others whose views do not agree with either of us. Everything depends upon what weight one gives to the number and disposition of the roots, as compared with the disposition of the cusps and their interpretation.

It appears that Robinson, from his study of the cast, is clearly not aware that what he refers to as "the ridge" linking the two cusps is divided by a very clear fissure, and is not a continuous ridge at all. This fissure shows very clearly in the photograph which I published and which was taken of the original tooth, but does not show in the photograph used by Robinson, which is apparently taken of a cast.

Finally, let me say that since the molar teeth of *Zinjanthropus* exceed the molar teeth of *Australopithecus* or *Paranthropus* in size, it is not unlikely that the deciduous molars of a descendant of *Zinjanthropus* should also be much larger than the deciduous molars of the known South African Australopithecinae, and since Chellean Stage I is a direct derivative of the Oldowan culture, it is reasonable to expect that the makers of Chellean Stage I will prove to be more advanced descendants of *Zinjanthropus*; heading in the *Homo* direction.

L. S. B. LEAKEY

Coryndon Museum,
Nairobi,
Kenya.

FORTHCOMING EVENTS

*(Meetings marked with an asterisk * are open to the public)*

Monday, February 8

British Society for the Philosophy of Science (in the Joint Staff Common Room, University College, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. R. L. Goodstein : "The Significance of Incompleteness Theorems".

Institution of Electrical Engineers, Electronics and Communications Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Discussion on "Present Views on Ground-Wave Propagation" opened by Mr. G. Millington.

University of London (at Senate House, London, W.C.1), at 5.30 p.m.—Prof. A. T. Ippen (Massachusetts Institute of Technology) : "Tide-dynamics in Estuaries".* (Further lectures on February 10 and 12.)

Royal Institute of Chemistry (joint meeting with the Sir John Cass College Chemical Society, at the Sir John Cass College, Jewry Street, London, E.C.3), at 6.30 p.m.—Prof. E. D. Hughes, F.R.S. : "Reaction Mechanisms".

Royal Geographical Society (at 1 Kensington Gore, London, S.W.7), at 8.30 p.m.—Mr. H. J. B. Harding : "The Channel Tunnel".

Tuesday, February 9

Zoological Society of London (at the Zoological Gardens, Regent's Park, London, N.W.1), at 5 p.m.—Scientific Papers.

Textile Institute (at the Chemical Society, Burlington House, Piccadilly, London, W.1), at 6.30 p.m.—Mr. E. Roberts and Mr. F. P. Thompson : "Domestic Washing of Textiles".

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Mr. E. C. Carter : "Model Testing in the Aircraft Research Association".

Radar and Electronics Association (at the Royal Society of Arts, John Adam Street, Adelphi, London, W.C.2), at 7.30 p.m.—Mr. E. Wolfendale : "Transistors—To-day and To-morrow".*

Wednesday, February 10

Physical Society, Low Temperature Group (at the Royal School of Mines, Prince Consort Road, London, S.W.7), at 4 p.m.—Dr. H. M. Rosenberg : "Some Low Temperature Experiments on the Alkali Metals".

Geological Society of London (at Burlington House, Piccadilly, London, W.1), at 5 p.m.—Sir Edward Bailey and Dr. W. J. McCallien : "Some Aspects of the Steinmann Trinity—Mainly Chemical" ; Dr. M. J. Fleuty : "The Three-Fold Systems in the Metamorphic Rocks of Upper Glen Orrin, Ross-shire and Inverness-shire" (to be read by Prof. J. Sutton).

Institution of Electrical Engineers, Supply Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. R. L. Grimsdale and Mr. P. H. Sinclare : "The Design of Housing-Estate Distribution Systems using a Digital Computer" ; Mr. U. G. W. Knight : "The Logical Design of Electrical Networks using Linear Programming Methods".

Society of Chemical Industry, Food Group—Nutrition Panel (at 14 Belgrave Square, London, S.W.1), at 6.15 p.m.—Mrs. P. Fisher and Miss M. Corbridge : "The Public Attitude to Nutrition".

Institute of Metals (at 17 Belgrave Square, London, S.W.1), at 6.30 p.m.—Dr. W. C. Winegard (University of Toronto) : "Solidification of Alloys".

Institute of Wood Science (at Imperial College, Exhibition Road, London, S.W.7), at 6.45 p.m.—Mr. G. Garlick : "The Quality and rading of Softwood in Relation to End Use".

British Interplanetary Society (in the Hoare Memorial Hall, ch House, Great Smith Street, Westminster, London, S.W.1), .m.—Prof. Alla Masevich : "Soviet Space Research".

ty for Psychical Research (at Kensington Town Hall, n High Street, London, W.8), at 7 p.m.—Mr. G. Zorab : hips in Spontaneous Paranormal Phenomena".

Engineering Society, London Branch (at Hope House, ter Street, Westminster, London, S.W.1), at 7 p.m.—Ball : "Uses of New Metals".

of Clinical Biochemists, Southern Region (in Lecture Theatre of the new Middlesex Hospital Medical , Cleveland Street, London, W.1), at 7.30 p.m.—Dr. : "Pitfalls in the Investigation of Metabolism through Some Cautionary Examples from Recent Studies of , Schizophrenia and Hartnup Disease".

Thursday, February 11

y (at Burlington House, Piccadilly, London, W.1), iss Elsie M. Widdowson and Prof. R. A. McCance, ffects of Accelerating Growth. I : General Somatic Mr. J. W. T. Dickerson and Miss Elsie M. Widdowects of Accelerating Growth. II : Skeletal Develop- Hicken and Mr. P. L. Krohn : "The Histocompatnts of Ovarian Grafts in Mice".

ty of London (at Burlington House, Piccadilly, 5 p.m.—Prof. C. F. A. Pantin, F.R.S. : "Alfred nd his Pre-Darwinian Essay" ; Mr. Quentin Bone : the Chordates", with a Discussion to be opened by r.

Antiquarian Horological Society (at the Science Museum, South Kensington, London, S.W.7), at 7 p.m.—Mr. B. Caillard : "Pendulum Watches".

Society of Instrument Technology (at Manson House, 26 Portland Place, London, W.1), at 7 p.m.—Mr. T. A. Lucas : "Measurement, Automatic Control and Data Reduction as Applied to a Cyclic Plant".

Chemical Society (in the Lecture Theatre, The Royal Institution, Albemarle Street, London, W.1), at 7.30 p.m.—Prof. P. L. Pauson : "Hydrocarbon-metal Carbonyls" (Tilden Lecture).

Friday, February 12

Association of Applied Biologists (at the Royal Entomological Society, 41 Queen's Gate, London, S.W.7), at 10.50 a.m.—Meeting on "Aspects of Crop Physiology".

British Psychological Society, Occupational Psychology Section (in the Department of Psychology, Birkbeck College, Malet Street, London, W.C.1), at 1 p.m.—Annual Meeting. Mr. D. E. Broadbent : "Industrial Inspection and Recent Advances in Psycho-Physics" (Chairman's Address).

University of London (at Bedford College, Regent's Park, London, N.W.1), at 5.30 p.m.—Prof. E. Wegmann (Neuchâtel) : "Some Aspects of the Geology of the Basements".*

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Mr. N. D. Norman and Mr. T. Clutterbuck : "Choice of Aerial Vehicle".

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Mr. F. T. Bacon : "Fuel Cells : Will They Soon Become Major Sources of Electrical Energy ?"

Saturday, February 13

Biochemical Society (at the Royal College of Surgeons, Lincoln's Inn Fields, London, W.C.2), at 10 a.m.—Symposium on "The Biochemistry of Mucopolysaccharides".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned :

Assistant (honours agricultural science graduate) in Crop Husbandry—The Secretary, Faculty of Agriculture, University College, Glasnevin, Dublin (February 12).

Lecturer in the Department of Mathematics—The Secretary, The University, Edinburgh (February 13).

Senior Lecturer or Lecturer in the Department of Electrical Engineering (Electronics)—The Registrar, The University, Liverpool, quoting Ref. CV/N (February 13).

Lecturer or Senior Lecturer in Biology ; and a Lecturer in Chemistry in the Training Department—The Warden, University of London Goldsmiths College, London, S.E.14 (February 15).

Plant Cytogeneticist, Scientific Officer (with an honours degree in botany, and preferably postgraduate experience in cytological research) at the Royal Botanic Garden, Edinburgh, for the cytological exploration of the living collections in the Garden and the development of the cytogenetic contribution to problems of plant taxonomy—The Establishment Officer (B), Ministry of Works, 122 George Street, Edinburgh 2 (February 15).

Senior Research Assistant (with a Ph.D. in theoretical physics or equivalent research qualifications, and preferably some experience in many body problems, or the methods of field theory) to Dr. N. H. March in the Department of Physics, for work concerned with electron correlation in metals—The Registrar, The University, Sheffield 10 (February 15).

Assistant Experimental Officer or Experimental Officer (graduate) in the Plant Pathology Department, for work on insect transmission of viruses—The Secretary, Rothamsted Experimental Station, Harpenden, Herts (February 20).

Assistant in Bacteriology (Science)—The Secretary of University Court, The University, Glasgow (February 20).

Demonstrator in Geography at the University of New England, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (February 20).

Lecturer and an Assistant Lecturer in Zoology—The Registrar, The University, Nottingham (February 20).

Lecturer in Philosophy ; a Lecturer in Social Anthropology ; and an Assistant Lecturer in Geography—The Secretary, The Queen's University, Belfast (February 20).

Lecturer or Assistant Lecturer in the Department of Applied Mathematics—The Registrar, The University, Liverpool (February 22).

Lecturer (preferably with an interest in biophysics) in Physics—The Secretary, St. Mary's Hospital Medical School (University of London), Paddington, London, W.2 (February 22).

Assistant Lecturer in Mathematics (Pure or Applied)—The Principal, Royal Holloway College (University of London), Englefield Green, Surrey (February 25).

Principal Lecturer and a Senior Lecturer in the Department of Mechanical Engineering ; Senior Lecturers (3) in the Department of Electrical Engineering ; a Lecturer and an Assistant Lecturer in the Department of Applied Physics ; a Senior Lecturer in Metallurgy and a Lecturer in Inorganic Chemistry in the Department of Chemistry, Metallurgy and Textiles—The Registrar, Lanchester College of Technology, Butts, Coventry (February 25).

Lecturer (with a good honours degree in electrical engineering or physics, and suitable experience in light-current electrical engineering and applied electronics) in Electrical Engineering—Joint Clerk to the University Court, Queen's College (University of St. Andrews), Dundee (February 27).

CHAIR AND HEADSHIP OF THE DEPARTMENT OF ZOOLOGY, and CHAIR AND HEADSHIP OF THE DEPARTMENT OF MINING—The Registrar, The University, Leeds 2 (February 29).

LECTURER or ASSISTANT LECTURER IN BIOCHEMISTRY—The Registrar, The University, Leeds (February 29).

LECTURER (with experience in archæological field work, and qualified particularly in British archæology) IN ARCHÆOLOGY—The Secretary and Registrar, University College of North Wales, Bangor, North Wales (February 29).

SCIENTIFIC OFFICERS/SENIOR SCIENTIFIC OFFICERS (analytical or inorganic chemists with a first- or second-class honours degree in chemistry or equivalent) IN THE LABORATORY OF THE GOVERNMENT CHEMIST, D.S.I.R., London, for analytical work including research into the development of advanced methods of analysis—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. 487/9A (February 29).

RESEARCH ASSISTANTS (2) (with a first- or second-class honours degree in chemistry or Graduate Member of the Royal Institute of Chemistry) IN CHEMISTRY—The Registrar, Lanchester College of Technology, Butts, Coventry (March 1).

RESEARCH FELLOW IN CRYSTALLOGRAPHY (COMPUTING) in the School of Chemistry, to take charge of the coding and supervise the use of computer programmes for crystallographic calculations of the high-speed 'Silliac' computer—The Registrar, University of Sydney, Sydney, N.S.W., Australia (March 4).

IMPERIAL CHEMICAL INDUSTRIES RESEARCH FELLOW IN CHEMISTRY OR PHYSICS—The Registrar, The University, Leicester (March 5).

IMPERIAL CHEMICAL INDUSTRIES RESEARCH FELLOWS IN BIOCHEMISTRY, CHEMISTRY, ENGINEERING AND PHYSICS—The Secretary and Registrar, The University, Southampton (March 5).

LECTURER IN APPLIED MATHEMATICS—The Registrar, King's College (University of Durham), Newcastle-upon-Tyne (March 5).

ASSISTANT LECTURER (veterinary surgeon, or with a degree in physiology) IN THE DEPARTMENT OF PHYSIOLOGY—The Secretary, Royal Veterinary College (University of London), Royal College Street, London, N.W.1 (March 15).

LECTURER or ASSISTANT LECTURER IN BOTANY at the University of Malaya (Singapore Division)—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 15).

CHAIR OF ZOOLOGY in the Imperial College of Tropical Agriculture, Trinidad, West Indies—The Secretary, Imperial College of Tropical Agriculture, 40 Norfolk Street, Strand, London, W.C.2 (March 17).

LECTURER (qualified in either pure or applied mathematics) IN MATHEMATICS—The Secretary of University Court, The University, Glasgow (March 19).

ASSISTANT LECTURER or LECTURER (with a strong interest in some branch of theoretical physics, or in information theory, or some other branch of applied mathematics) IN APPLIED MATHEMATICS—The Registrar, University College of South Wales and Monmouthshire, Cathays Park, Cardiff (March 21).

LECTURER or ASSISTANT LECTURER IN ZOOLOGY at the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, March 21).

LECTURERS IN PURE MATHEMATICS—The Secretary, The Queen's University, Belfast (March 21).

LECTURER (preferably with interests in theoretical electromagnetics and/or plasma theory) IN PHYSICS—The Secretary, The Queen's University, Belfast (March 30).

CHAIR OF BOTANY in the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, March 31).

ASSISTANT LECTURER (Grade "B") TO TEACH ORGANIC AND GENERAL CHEMISTRY—The Clerk to the Governors, Northern Polytechnic, Holloway, London, N.7.

ASSISTANT LECTURER IN BOTANY; an ASSISTANT LECTURER IN CHEMISTRY; an ASSISTANT LECTURER IN PHYSICS; an ASSISTANT LECTURER IN PHYSIOLOGY; and a RESEARCH ASSISTANT (qualified in biochemistry, physiology or zoology) TO THE PROFESSOR OF ZOOLOGY—The Secretary, Queen Elizabeth College (University of London), Campden Hill Road, London, W.8.

ELECTRONICS ENGINEER (Experimental Officer grade), to collaborate with research and instrument development in marine physics at Millport—The Secretary, Scottish Marine Biological Association, Millport, Isle of Cumbrae, Scotland.

EXPERIMENTAL OFFICER (preferably with a pass degree or H.N.C., and experience in electronic and optical recording techniques, or G.C.E. in five subjects, including two scientific or mathematical subjects at "A" level), to work in collaboration with scientists on shock and detonation wave phenomena using high speed electronic and optical equipment—The Senior Recruitment Officer, The Atomic Weapons Research Establishment, Aldermaston, Berkshire, quoting Ref. A.2366/34.

LECTURER IN MATHEMATICS; and a LECTURER IN STATISTICS AND MATHEMATICS—The Registrar, Bradford Institute of Technology, Bradford 7.

MASTER TO TEACH PHYSICS to Open Scholarship level—The Headmaster, Latymer Upper School, Hammersmith, London, W.6.

MASTER TO TEACH PHYSICS to Sixth Form standard—The Headmaster, Merchant Taylors' School, Sandy Lodge, Northwood, Middlesex.

SENIOR LECTURER (with a good honours or higher degree, with teaching and preferably appropriate industrial experience) IN THE DEPARTMENT OF CHEMISTRY AND METALLURGY, to teach inorganic chemistry to final B.Sc. and Part II Grad.R.I.C. level—The Principal, Rutherford College of Technology, Northumberland Road, Newcastle-upon-Tyne.

SENIOR TECHNICIAN IN THE BIOCHEMISTRY DEPARTMENT—The Secretary, Queen Mary's Hospital for the East End, Stratford, London, E.15.

SENIOR TECHNICIAN (preferably man 25–30 years of age, with biochemical and practical laboratory experience) for newly formed Research Department of Immunology—The Secretary, Wright-Fleming Institute, St. Mary's Hospital Medical School, Paddington, London, W.2.

REPORTS and other PUBLICATIONS

(*not included in the monthly Books Supplement*)

Great Britain and Ireland

Department of Scientific and Industrial Research. Food Investigation. Technical Paper No. 7: Quality Control in Marketing Fresh Poultry. By Dr. D. H. Shrimpton. Pp. iv+25. (London: H.M. Stationery Office, 1959.) 2*s*. net. [312

Eton College Natural History Society. Fourteenth Report, 1958–1959. Pp. 35+2 plates. (Eton: Eton College Natural History Society, 1959.) 2*s*. 6*d*. [312

Department of Scientific and Industrial Research: Building Research Station. National Building Studies Special Report. No. 30: A Study of Alternative Methods of House Construction. Pp. vi+93+19 plates. (London: H.M. Stationery Office, 1959.) 7*s*. 6*d*. net. [812

Department of Scientific and Industrial Research. Pest Infestation Research—Bulletin No. 1: Fumigation with Methyl Bromide Under Gas-Proof Sheets. By W. Burns Brown. Pp. vi+44+8 plates. (London: H.M. Stationery Office, 1959.) 4*s*. 6*d*. net. [812

Commonwealth Mycological Institute. Mycological Papers. No. 71: Studies on Cercospora and Allied Genera. I: *Cercospora* Species with Coloured Spore on *Phyllanthus* (Euphorbiaceae). By F. C. Deighton. Pp. 23. 9*s*. net. No. 72: Clasterosporium and Some Allied Dematiaceae—Phragmosporæ. II. By M. B. Ellis. Pp. 75. 25*s*. net. (Kew: Commonwealth Mycological Institute, 1959.) [812

Other Countries

United States Department of the Interior: Geological Survey. Bulletin 1039–C: Haydite Raw Material in the Kings River, Sutton, and Lawing Areas, Alaska. By Richard A. Ackhart and George Plafker. Pp. iv+33–65+plates 7–10. Bulletin 1046–Q: Stratigraphy of Triassic and Associated Formations in Part of the Colorado Plateau Region. By John H. Stewart, George A. Williams, Howard F. Albee, and Omer B. Raup. With a section on Sedimentary Petrology by Robert A. Cadigan. Pp. vi+487–576+plate 49. 65 cents. Bulletin 1052–I: Equation in Continuity in Geology with Applications to the Transport of Radioactive Gas. By A. Y. Sakakura, Carolyn Lindberg and Henry Faul. Pp. iv+287–305. 15 cents. Bulletin 1058–G: Geology of the Mount Katmai Area, Alaska. By A. Samuel Keller and Hillard N. Reiser. Pp. iv+261–298+plates 29–32. Bulletin 1072–I: Beryl Deposits of the Beecher No. 3-Black Diamond Pegmatite, Custer County, South Dakota. By Jack A. Redden. Pp. iii+537–559+plates 17 and 18. Bulletin 1084–D: Geochemistry of Uranium in Phosphorates and Black Shales of the Phosphoria Formation. By Richard P. Sheldon. Pp. iv+83–115. 20 cents. Bulletin 1087–B: Uranium Resources of the Cedar Mountain Area, Emery County, Utah—A Regional Synthesis. By Henry S. Johnson, Jr. Pp. iv+23–58. 20 cents. (Washington, D.C.: Government Printing Office, 1959.) [312

Berichte des Deitschen Wetterdienstes. Nr. 57 (Band 8): Ein Beitrag zu den Windverhältnissen in den ersten 100. der Atmosphäre. Von Rüdiger Trappenberg. Pp. 41. (Offenbach a.M.: Deutschen Wetterdienstes, 1959.) [812

Smithsonian Institution: United States National Museum. Bulletin 218: Contributions from the Museum of History and Technology, Papers 1 to 11. Pp. v+201. (Washington, D.C.: Government Printing Office, 1959.) 2.50 dollars. [812

National Academy of Sciences—National Research Council, Washington. Publication No. 713: Digest of Literature of Dielectrics, Vol. 22, 1958. Prepared by the Committee on Digest of Literature of the Conference on Electrical Insulation, Division of Engineering and Industrial Research. Edited by Robert A. Soderman and Louis J. Frisco. Pp. xiii+293. (Washington, D.C.: National Academy of Sciences—National Research Council, 1959.) 5 dollars. [812

Smithsonian Miscellaneous Collections. Vol. 139, No. 8: The Anatomical Life of the Mosquito. By R. E. Snodgrass. Pp. iii+87. (Publication 4388.) (Washington, D.C.: Smithsonian Institution, 1959.) [812

United States Department of Agriculture: Agricultural Research Service. Technical Bulletin No. 1203: Measuring Seepage fror Irrigation Channels. By A. R. Robinson and Carl Rohwer. (Publish in co-operation with the Colorado Agricultural Experiment Station the Bureau of Reclamation, United States Department of the Inte Pp. ii+82. (Washington, D.C.: Government Printing 1959.)

Proceedings of the United States National Museum. No. 3419: A Description and Classification of the Final Ins of the Ichneumonidæ (Insects, Hymenoptera). By J. R Pp. 391–511. (Washington, D.C.: Government Prin 1959.)

Comité International de Photobiologie. Bulletin Nc 1959. Pp. 19. (Copenhagen: Dr. Bent Buchmann, Fir Hospital, Strandboulevarden 49, 1959.)

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EVOLUTION OF MANKIND

IN his speech opening the debate on science in civil life in the House of Lords on December 9, Lord Taylor, in referring to the vanishing difference between pure and applied science, quoted both from Lord Adrian's recent Fawley Lecture on November 17 to the University of Southampton on "The Risks of Progress" and from Prof. P. B. Medawar's Reith Lecture on "The Future of Man". Lord Adrian remarked that we have become more and more aware that concentration on practical issues may lead to big theoretical advances ; and he added that there is no longer any stigma attached to an idea or technique derived from a workshop rather than a university. Lord Taylor observed that applied science might lead to unexpected fundamental discovery, and pure science might yield utilitarian results when least intended ; nevertheless, the difference remains, and in applied science someone must indicate the objective.

Considering more particularly medical science, Lord Taylor referred to the growing importance of producing health and fitness, but he used the term fitness in a rather wider sense than that of 'net reproductive advantage' used by Prof. Medawar in his second Reith Lecture, though Lord Taylor did not deny the importance of this definition in long-term genetic study. In this second lecture, Prof. Medawar considered the question whether man could go on evolving in the future as he has evolved in the past, or whether there is a good reason why his evolution should now have come to an end, and then asked, as a subsidiary question, whether there is any real reason for supposing that advances in medicine and hygiene are undermining the fitness of the human race. It is in this context that Prof. Medawar uses the word 'fitness' in the sense of 'net reproductive advantage', as it is generally used by students of heredity, though he recognizes that it neglects much that is deeply important in human life.

It may be recalled that in his first lecture, Prof. Medawar had emphasized the fallibility and limitations of our efforts to predict the future. Nevertheless, he maintained in his second lecture that it is not true to say that advances in medicine and hygiene must cause a genetical deterioration of mankind ; there is more to be feared from a slow decline of human intelligence. But if that is happening, it is because the rather stupid are biologically fitter than those who are innately more intelligent, not because medicine is striving to raise the biological fitness of those people who might otherwise be hopelessly unfit.

This relation between intelligence and fertility was discussed more particularly in his fifth lecture, in which Prof. Medawar pointed out that while the average level of intelligence may in fact decline, this is no more than a presumption, first because one highly important piece of information, about the intelligence of married couples who have no children, or of people who never marry at all, is missing, and secondly, because we do not yet know exactly how differences of intelligence are inherited. Moreover, profound changes in fertility have been taking place during the past fifty or a hundred years, and they are not yet complete. Any decline of intelligence may be a purely temporary phenomenon, marking the transition from free reproduction accompanied by high mortality to restricted reproduction accompanied by low mortality.

There may thus be less difference between Prof. Medawar's views and that expressed by Lord Adrian in his Fawley Lecture than appears at first sight, when we contrast Prof. Medawar's words with Lord Adrian's balance sheet. Lord Adrian, too, emphasizes the need for much more information, and he himself suggests that we need not be too downcast at the picture he presented of the result of indiscriminate life-saving. We have succeeded so well, he observes, in our aim at keeping alive every child that is born that we are certainly preserving many unfavourable genes which would otherwise have died out. "If we set out to save the unfit we must expect more unfitness in the world and more inheritance of the factors which promote it. Even if the radiation level remains as it now is, the advance of science can harm the genetic constitution of the race".

Lord Adrian goes on to point out that even a doubling of the mutation-rate, however, would not double the load of inherited weakness until many generations had elapsed, for many of the faulty genes will be recessive or sex-linked. In the next thirty years we can confidently expect a great increase in our understanding of the ultimate structure of living matter, of viruses and chromosomes and of the whole molecular mechanism of inheritance and growth. While it would be idle to look further ahead than one generation, Lord Adrian is confident that at the present rate of progress the medical science of our great-grandchildren will make them far more capable of preserving the race. Moreover, Prof. K. Mather, in a recent broadcast on heredity and environment, has stressed the way in which human ideas and the power to record and communicate them make our social evolution a far more rapid process than our biological evolution. The effects of new knowledge take far less time to be established than those of a new mutation, and while the prospect of decline in the level of human ability is far more disturbing than that of a rise, Lord Adrian believes that our increased knowledge will enable us to find a way to escape the genetic dangers which may threaten us before our stock has been seriously harmed.

That, however, does not give a fair picture of Lord Adrian's approach to this question of the risks of progress. He began by directing attention to the way in which academic science and its relation to technology and industry have been violently forced out of the pattern which T. H. Huxley prescribed for it in the Victorian period. We have become painfully aware of the risks we may be running in allowing free play to all the developments of science in the technological field, and while most of us would be shocked at the suggestion of banning any attempt to pursue a line of scientific inquiry which aimed primarily at increasing our understanding of the natural world, most of us would cheerfully agree to the prohibition of work on certain practical developments if we thought they might have a harmful effect on the human race.

It is at this point that Lord Adrian brought this distinction between pure and applied science sharply into focus on the problems of the under-developed territories of the world. Much remains to be done before those territories can be freed from poverty and hard labour; but the objective cannot be attained if the timing is not adequately supervised, if the population of the world is allowed to increase more rapidly than we can increase our means for feeding and housing it, if we exhaust our stores of metals, coal and oil before we have solved the problem of fusing hydrogen atoms. Similarly, disaster may follow if we press ahead too rapidly along particular lines in particular parts of the world without reference to the general welfare, and our distinction between primary and secondary needs must take increasing account of the human requirements in increased leisure. The risks of progress, Lord Adrian urged, should be calculated risks, based on two principles: our concern about the risk of serious disease rather than the risk of injury, and our concern that the human race should remain at least as healthy and intelligent as it is now.

Lord Adrian illustrated his point in a general way by referring to the radiation hazard. Thus he remarked that there will be many countries using nuclear power which may be less careful than ourselves in the disposal of radioactive wastes, and in seeing that fires and explosions do not generate clouds of radioactive material. Precautions taken in Britain will not necessarily protect us from the results of large-scale disasters elsewhere, and it is especially in the context of the aid to the under-developed countries that we should begin, as Lord Adrian suggested, to consider what risks a civilized community should regard as reasonable to accept. Indeed, as should already be apparent, the present risks which this age confronts arise largely out of the disparities which exist in the societies which constitute civilization, whether within or across national frontiers, and the stresses which such disparities in standards of living or in the capacity for adaptation and assimilation of technical advance are liable to cause. With the increasing attention now being focused on Western aid to the under-developed countries, either technical or economic, it is the more important that due thought should be given to this aspect, and that technical assistance should be considered realistically and impartially in its widest context.

STELLAR POPULATIONS

Stellar Populations

Proceedings of the Conference sponsored by the Pontifical Academy of Sciences and the Vatican Observatory, May 20–28, 1957. Edited by D. J. K. O'Connell, S.J. Pp. lxv + 544. (Castel Gandolfo: Specola Vaticana, 1958. Distributed by North Holland Publishing Company, Amsterdam; and Interscience Publishers, Inc., New York.) 76*s*.

THIS is not a text-book; neither is it merely a collection of independent papers on a common theme. It is the report of a successful conference held at the Vatican Observatory in 1957 on the complicated and rapidly changing problem of stellar populations.

The fundamental questions are how and when were the stars formed, how did the Milky Way come into being and what has determined its structure as we see it to-day? If we could sort out all the stars around us into groups or populations, according to age and origin, we should fairly certainly be much further on than now in our understanding of these larger questions. Only fifteen years have passed since Baade announced his now famous hypothesis that all stars belong to one of two families: Population I, the members of which are comparatively young, and are associated with interstellar matter and the spiral arms of galaxies; Population II, comparatively old and to be found in places where interstellar matter is scarce or non-existent, for example, the globular clusters, the elliptical galaxies, the nuclei of spiral nebulæ. Since then general opinion has come gradually to the view that the concept of two clear-cut populations is an over-simplification, and a good deal of time at this conference was taken up with producing a more acceptable scheme, not too elaborate in relation to the still very elementary nature of our knowledge.

The reader of "Stellar Populations", if he is to get much from the book, needs some idea of the underlying thoughts, not very explicitly described, which form the background of such discussions to-day. It is unlikely that all the participants had exactly the same picture in mind, and perhaps it is rash to attempt to put it into words. Although no fairy story, it could well start off with the familiar phrase 'once upon a time'; for the time-scale of the earlier stages of the Milky Way's history is still extremely uncertain.

Anyway, at this remote epoch there is supposed to have been a primitive diffuse gas of practically pure hydrogen. Somehow or other, by processes we do not properly understand, this gas condensed into first-generation stars (Baade's Population II), and at or about the same time the stars and gas condensed into a Milky Way system, not necessarily closely similar to our present Milky Way. This may have happened 5–10 thousand million years ago. The more massive of the first generation stars evolved rapidly, 'burned' their hydrogen into helium and later their helium into heavier elements. Theory shows us how this could happen, although only the fringe of the problem of computing the life-histories of appropriate star models has yet been tackled, but very significantly we have here a possible mechanism for the origin of the heavier elements. Some of the more rapidly evolving stars became unstable and either by catastrophic explosions or by shedding material from their outer layers (both processes can be observed to-day) 'enriched' the interstellar gas with helium and heavier elements as well as hydrogen. Afterwards there was more star-building from the interstellar gas. We are uncertain when this occurred, whether during critical periods when conditions were specially favourable, or at a more or less steady rate throughout the history of the Milky Way. We feel confident that some star formation has continued until astronomically very recent times, 1–10 million years ago (for Baade's Population I), and is probably still continuing to-day. During this history of perhaps 10^{10} years, the Milky Way evolved into the system we are now trying to explore by various techniques: a thin, flat, slowly rotating disk of stars and interstellar matter, with some spiral structure associated with dust, gas and young stars, and a more uniform background of older stars. The younger stars and the interstellar gas rotate in approximately circular orbits about the centre of the system; older stars tend to follow more elongated orbits, roughly elliptical, taking them at times near the centre. Around the centre older stars predominate and the system is supposed to have a nucleus and a central bulge similar to those we see in many external galaxies. Our picture includes also a very low-density halo, mainly gaseous but with a few stars, roughly spherical and for the most part detectable only by radio methods. The connexion of this halo with the rest of the system is so far not at all clearly understood.

Our ideas as to how all this came about are in detail till very vague. It is not unlikely that some essential arts of the problem have still eluded us, but so many different lines of evidence tend to point in the ame general direction that we are persuaded that oughly we are thinking along the right lines. The roblem of stellar populations is to sort out the stars as we see them to-day into some kind of an age order, and then to study their other physical properties in relation to this. We try to do this by using positions in the H–R. diagram, velocities, colour characteristics, or spectra, depending on what is available. The measurements are generally tedious, they often need large telescopes, and even when obtained the results may be inconclusive or ambiguous. Theoreticians have tasks as formidable as those of observers. For example, one of the conference members (Hoyle) estimated that something of the order of 10,000 hr. would be necessary on the largest existing electronic computers, to calculate the life-histories of a reasonably representative set of star models. Clearly, we are unlikely to get quick answers to any of the bigger questions, and since this kind of astronomical problem usually grows in complexity as our knowledge increases, the final answers may yet be a very long way off.

Meanwhile, here is a picture of the subject as it was in 1957, not a neat text-book account for a beginner, but a report of research in progress, with untidy ends everywhere, full of food for thought, but also capable of stirring up dissatisfaction, even irritation, in a critical reader. The organizers of this conference had the wisdom to keep it small. Among other great advantages this kept the discussions informal, spontaneous, well-informed and relevant. The reporting of both papers and discussions appears to have been done excellently.

R. O. Redman

HISTORY OF SCIENCE

A Short History of Scientific Ideas to 1900

By Dr. Charles Singer. Pp. xviii+525. (Oxford: Clarendon Press; London: Oxford University Press, 1959.) 35*s*. net.

WHEN Dr. Singer's "Short History of Science to the Nineteenth Century" appeared in 1941 it was the first, in the English language and within a reasonable compass, to "present, in simple form, the development of the conception of a rational and interconnected material world"; to relate the particular activity we call 'science' to the remaining intellectual activities of the heirs of Greco-Roman civilization. To emphasize the progressively changing character of that relationship the author introduced such leading phrases as "The Great Adventure" (Athens), "The Failure of Nerve" (Alexandria), "The Failure of Inspiration" (Rome), "The Failure of Knowledge" (Middle Ages), "The Revival of Learning", "The Insurgent Century", and "The Mechanical World". The present version—Dr. Singer rightly claims that it is "much more than a revision and rather more than a new edition"—contains in addition a succinct account (contributed by Dr. Angus Armitage) of the earliest records of systematized knowledge in the Middle East, and an additional Section—"Culmination of the Mechanical View of the World"—running to nearly 100 pages, rather less than half of which have been written by Prof. H. Dingle.

The book has been further enriched by two contributions by Dr. Derek Price, one on the work and instruments of Ptolemy, the other on the plane astrolabe. A few sub-sections, notably one on the "Status of Medieval Science", have been added, and others (for example, on the sixteenth century herbalists and anatomists) largely re-written. The importance of technological change is everywhere brought into greater prominence, and this is reflected among the much larger number of admirable illustrations, for the use of many of which, from "A History of Technology", Dr. Singer expresses gratitude to the Directors of Imperial Chemical Industries, Ltd.

The original "Short History", admirable as it was, could perhaps be criticized on the grounds of its sweeping condemnation of medieval science as a "period of intellectual degradation" (p. 161); the lack of balance in respect of the history of chemistry; and but a scant reference to embryology. Whereas the last lack has now been made good, it seems regrettable that, with the change of title and with the wealth of studies on the medieval period since the first appearance of the earlier version, the opportunity was not taken to present the medieval architects of ideas in a more sympathetic light. In regard to chemistry the situation has actually deteriorated. Though Prof. Dingle graciously allows that "the history of physics for a long time to come will not be without a chemical chapter" his ingeniously phrased "oraison funèbre", though not without expressions of profound respect for the departed, will scarcely pass as 'history'. Frankland did not just "assign" to each atom a property called valency; he was driven to speak (rather apologetically) of a "saturation capacity" in consequence of results unexpectedly obtained in his efforts to isolate a compound radical. To ignore the paths by which the Pisgah-views, now shared and interpreted by theoretical physicists, have been reached is to nullify a great deal of the cultural value of the history of science.

The faults, however, cannot outweigh the delight which awaits the reader in the many wise comments and illuminating comparisons scattered throughout the whole book. Perhaps, when in a few years time a third version is called for, the master from whom we have all learned so much might consider the dropping of the catch-titles. These may have been justified when first introduced ; but has not the time now come when a history of science should place the emphasis on *wie es eigentlich gewesen* ?

WILLIAM P. D. WIGHTMAN

AGRICULTURAL RESEARCH IN INDIA

Agriculture and Animal Husbandry in India

By Dr. M. S. Randhawa. Pp. xvi + 364. (New Delhi : Indian Council of Agricultural Research, 1958.) Rs. 15.00.

Mechanical Cultivation in India

By D. A. Gadkary. Pp. ix + 147 + 10 plates. (New Delhi : Indian Council of Agricultural Research, 1957.) Rs. 7.25 ; 11*s*. 6*d*.

AGRICULTURAL research in India is a subject of such importance that it is scarcely an exaggeration to say that the future of the world may depend upon it. Indian scientists and the Indian Council of Agricultural Research have had quite a good record so far, though both the financial and human resources of India fall far short of the real requirements of the situation. As Dr. Randhawa freely admits, other countries faced with the problem of obtaining maximum food supply from a limited area—Japan, Italy and Egypt—obtain yields per acre three or four times India's. If India could raise her yields to these levels she could over-feed any conceivable population she is likely to have for a very long time to come. But mere transplantation of seeds, fertilizers and methods of cultivation from, say, Japan, will not suffice. Plant breeding, as optimistic farmers have found to their cost, is a laborious task which has to be done over again for even the slightest difference in climate, and then again as new pests appear.

"Agriculture and Animal Husbandry in India" does not deal with the problems on the actual frontier of research, though the compilers are clearly aware of them. It is a compendium of information on the best strains of plants now available, cultivation requirements, expected yields, and where the seeds can be obtained. The geographical maps and statistical tables have been well done, though the chapter on irrigation is disappointingly superficial, and livestock receive much less attention than crops.

For every scientist engaged in original agricultural research India will need perhaps a hundred men to disseminate his results, as well as the elements of agricultural science, among an uninformed people. A book like this should play a valuable part in their training, and be a useful reference book to librarians in other countries.

"Mechanical Cultivation in India" is not broad enough in its scope to constitute an economic study of the circumstances (admittedly limited) in which mechanical cultivation is now worth introducing in India. Nor is it detailed enough to serve as an operator's handbook. It appears to have been designed more as an administrative handbook, imparting the necessary minimum of information about types of machines, and making preparations for the administrative, recording and accounting duties which will arise if mechanical implements are to be operated from governmental tractor stations.

COLIN CLARK

INDUSTRIAL GUMS

Industrial Gums

Polysaccharides and Their Derivatives. Edited by Roy L. Whistler and James N. BeMiller. Pp. xi + 766. (New York : Academic Press, Inc. ; London : Academic Press, Inc. (London), Ltd., 1959.) 25 dollars.

BRIDGING the gap between the academic chemist and the chemical technologist is always a problem since in general their points of view differ widely. Here it has been achieved by competent academic editing of a series of chapters written mainly by industrial chemists. The brief 'academic' chemistry of gums included in the book does not interfere, and is indeed necessary, for the main task of presenting detailed information of those practical applications of gums which are of particular interest to food technologists, paint and textile manufacturers, paper industrialists and pharmaceutical chemists. The current prices and potential resources of man of the gums are included. Certain gums which, as ye have not been used widely in industry are include together with suggestions as to their possible applic tions.

My minor criticisms of this book include a lack consistency from one chapter to another in depicti of polysaccharide structures and indeed their co plete absence in certain chapters. In the otherwis competent chapter on dextran I could find no details of the Swedish manufacture of dextran or of the use of iron complexes of dextran. The author was also unaware that the structure of the mycodextran from *Aspergillus niger* (p. 552) had been extensively investigated and that it had been renamed nigeran, due to its marked structural difference from normal dextrans. The more detailed formula for laminaran postulated by Peat and his colleagues could well have been included. In the chapter on agar no mention is made of its application in the now widely used agar diffusion technique. The authors of the agar article are employed by the only American company manufacturing agar, and can perhaps be forgiven for advertising the superiority of their company's product as opposed to that of the much cheaper Japanese agar. The book gives detailed coverage of the literature up to the end of 1957 with scattered references from 1958.

I can thoroughly recommend this book as essential for all technical libraries, providing as it does comprehensive references to the numerous patents on the industrial uses of gums. I am sure it will be read more widely than its title suggests, since by analogy numerous applications can be suggested for many unexploited polymers, particularly as attempts are made throughout the book to relate the physical characteristics of the gums to their chemical structure. After editing this book, Prof. Whistler will be much sought after as an industrial consultant.

S. A. BARKER

Radioisotopes in Scientific Research

Proceedings of the International Conference held in Paris in September 1957, under the auspices of the United Nations Educational, Scientific and Cultural Organization. Edited by Prof. R. C. Extermann. Vol. 1: Research with Radioisotopes in Physics and Industry. Pp. xxi+761. Vol. 2: Research with Radioisotopes in Chemistry and Geology. Pp. xxi+741. (London and New York: Pergamon Press, 1958.) 140*s.* net each volume.

THE Unesco Conference on "Radioisotopes in Scientific Research" has been reported in *Nature*, **180**, 1029 (1957). The proceedings, which have now been published, comprise the contributions in their original language, but the abstracts of each paper are given in English, French, Russian and Spanish. The discussions are reported for each paper, and, although greatly reduced in volume, are well edited. Contrary to general belief that such research outdates itself in a reasonably short time, one has to admit that some of the material presented will be extremely useful to workers in this field for many years to come. For example, the work reported on the tracing of water by using cosmic-ray and man-made tritium, by Begemann and Libby, is only now fully appreciated, and work in years to come will be based on the ideas contained in this paper. The proceedings of this most useful conference should certainly be in every science library and, to a certain extent, they can be regarded as reference books.

Although the reproduction and printing are reasonable, if not in any way elaborate, the price of the volumes seems rather high. It is a pity that the book, which came out one year after the conference was held, still contains a number of misprints, the most misleading ones being found in the index of Vol. 2, where twenty-two contributions have been indexed with the wrong page number owing to the fact that one contribution was erroneously split in two, whereas at the end of the book the page number again agrees with that of the papers, because one page number was left out for one contribution altogether.

H. SELIGMAN

Nitrogen Metabolism in Plants

By Prof. George C. Webster. (Row–Peterson Biological Monographs.) Pp. viii+152. (Evanston, Ill.: Row, Peterson and Company, 1959.) 3.50 dollars.

THIS is an age in which authoritative reviews in the fields of plant physiology and plant biochemistry are tending to be concentrated in encyclopædias or other massive volumes which are too expensive for individual ownership; indeed, the influence of such compilations on scientific thought may prove to be limited because of the physical difficulty of consulting them. Thus, there is still very much room for smaller publications in which substantial fields of investigation are reviewed authoritatively and, though not exhaustively, with sufficient documentation for the reader to be made aware of the more important original sources. Such a review is provided by this very useful little book, which is concerned mainly with the chemical pathways of nitrogen metabolism in plants and has been written by an author who is already well known for his own original studies in this field. The topics covered are nitrogen nutrition, amino-acid metabolism, biosynthesis of amides and peptides, protein and nucleic acid metabolism, nitrogen metabolism of excised organs and ageing tissues, and translocation of nitrogenous substances. The book is written in straightforward and clear style, marred only occasionally by infelicitous phrases or repetitive treatment. In a few instances (for example, Table 2) the identity of the plant material to which the data presented refer is not stated; three misprints were noted, while an arrow appears to be missing from Fig. 26. These are minor blemishes in an otherwise workmanlike text, which I recommend for use by research workers and honours students.

G. BOND

Bird Watching for Beginners

By Bruce Campbell. (Puffin Book PS71.) Pp. 240. (Harmondsworth, Mddx.: Penguin Books, Ltd., 1959.) 2*s.* 6*d.*

THIS is a revised edition of a book first published in 1952. It is divided into three sections. The first explains how to become a bird watcher and the limits of classification. The second discusses two hundred common British resident and visiting species in systematic order from crows to game-birds. It deals mainly with relationships, distribution and habits, and points out aspects "suitable for study by the ordinary bird watcher".

Part 3 briefly surveys various problems; how to set about identification, how to make bird counts, how to study birds at the nest, and so on.

The author is highly qualified to give instructions to beginning bird watchers. He has an extensive practical knowledge of field-study and his association with the British Trust for Ornithology has given him a wide background of the subject based on the experience of others. Ability to impart this knowledge has been gained from long practice as lecturer and broadcaster. He has a readable style and pleasing personal approach which not only provides information but also inspires enthusiasm.

J. D. MACDONALD

The Rhondda Valleys

A Study in Industrial Development, 1800 to the Present Day. By E. D. Lewis. Pp. xiii+312+3 plates. (London: Phoenix House, Ltd., 1959.) 25*s.* net.

THE Rhondda became a borough in 1955 and to mark the event sponsored the publication of this excellent local history. Begun twenty years before as a master's thesis for the University of Wales, its author, the son of a Rhondda collier and now a Rhondda headmaster, has expanded his work to a scholarly and well-documented local history. The story of the Rhondda is well known, being that of a sparsely populated, picturesque, well-wooded valley, rarely penetrated by strangers at the beginning of the nineteenth century; it passed through a whole cycle of industrial development within the next hundred years, to be hit with more than average severity by the depression following the First World War. What would perhaps, in another continent and another context, have become a ghost town as its *raison d'être* faded, had too great a reservoir of human vitality to succumb. Much of the book is devoted to the human story—the early working conditions, the rise and temporary fall of trade unionism, and the later role of the Rhondda as stormy pioneer and leader in new concepts of management and labour relationships. The final chapter, dealing with the period from 1914 to the present day, makes it clear that the story is far from ended.

L. DUDLEY STAMP

STRUCTURE OF HÆMOGLOBIN

A THREE-DIMENSIONAL FOURIER SYNTHESIS AT 5·5-Å. RESOLUTION, OBTAINED BY X-RAY ANALYSIS

By DR. M. F. PERUTZ, F.R.S., DR. M. G. ROSSMANN, ANN F. CULLIS, HILARY MUIRHEAD and DR. GEORG WILL
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VERTEBRATE hæmoglobin is a protein of molecular weight 67,000. Four of its 10,000 atoms are iron atoms which are combined with protoporphyrin to form four hæm groups. The remaining atoms are in four polypeptide chains of roughly equal size, which are identical in pairs[1-3]. Their amino-acid sequence is still largely unknown.

We have used horse oxy- or met-hæmoglobin because it crystallizes in a form especially suited for X-ray analysis, and employed the method of isomorphous replacement with heavy atoms to determine the phase angles of the diffracted rays[4-7]. The Fourier synthesis which we have calculated shows that hæmoglobin consists of four sub-units in a tetrahedral array and that each sub-unit closely resembles Kendrew's model of sperm whale myoglobin[6]. The four hæm groups lie in separate pockets on the surface of the molecule.

Method of Analysis

Horse oxyhæmoglobin, crystallized from 1·9 *M* ammonium sulphate solution at *p*H 7, has the space group *C*2 with two molecules in the unit cell which lie on dyad axes[8]. In order to determine the phase angles of the 1,200 reflexions contained in the limiting sphere of 5·5 Å.$^{-1}$, six different isomorphous heavy-atom compounds were used (ref. 9 and unpublished work). Intensities were measured photographically and by counter spectrometer (Arndt, U. W., and Phillips, D. C., unpublished work). The relative positions and shapes of the heavy-atom replacement groups were found by correlation functions based on Patterson methods[10] and refined by least squares procedures. For each reflexion the structure amplitudes of all seven compounds were combined in an Argand diagram[11], and the probability of the phase angle having a value α was calculated for α = 0 10, . . . 355°. The centroid of the proba bution, plotted around a circle, was the the best vector *F* in the Fourier synth ...e results were finally plotted on 32 con ...r maps showing the distribution of electron density in sections spaced 2 Å. apart normal to *b* (Fig. 1). The absolute configuration of the molecule was determined from anomalous dispersion[4].

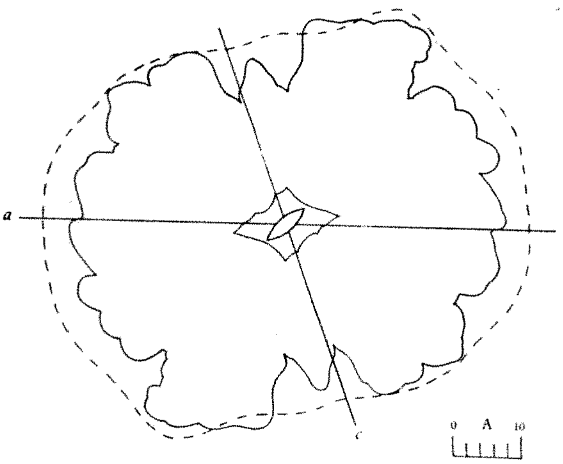


Fig. 2. External shape of the molecule. Full lines indicate the boundary derived from the contour at 0·54 electron/Å.^{3}. The broken line shows the boundary derived by Bragg and Perutz from two-dimensional data (ref. 13). Note the hole in the middle

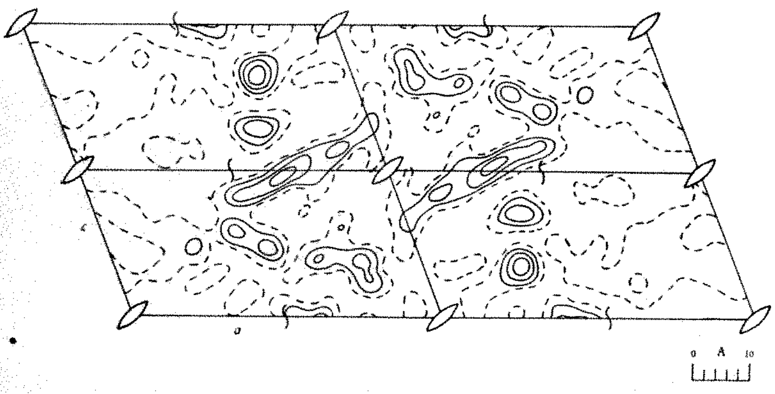
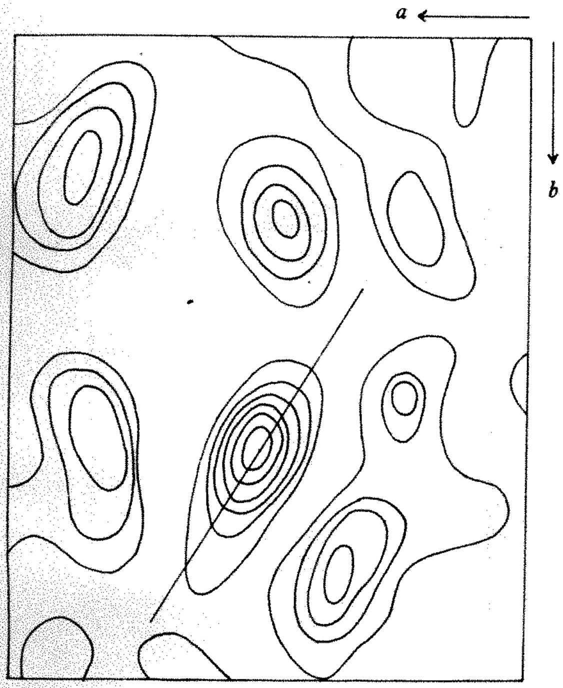


Fig. 1. Section at $y = 1/32b$. This cuts through the middle of the molecule on which the diagram is centred. 'Flat' areas indicating liquid appear on the left and right. Contours are drawn at intervals of 0·14 electron/Å.^{3}. The broken line marks 0·4 electron/Å.^{3}. Contours at lower levels are omitted

External Shape of the Molecule

More than half the volume of the crystals is taken up by liquid of crystallization, which mainly fills the spaces between the molecules and shows up on the contour maps in the form of flat, featureless regions (Fig. 1). The outlines of the two molecules in the unit cell can be traced by following the boundaries between these regions and the continuous electron-dense regions described below. In Fig. 2 the outline of one molecule, traced from the periphery of the 0·54



Hæm 1 and 2

Fig. 3. Section at $z = 1/4\ c$ showing one of the two hæm groups in the asymmetric unit. The straight line indicates the hæm orientation derived from electron spin resonance (ref. 14). The lowest contour shown is at 0·4 electron/Å.^{3}

electron/Å.^{3} contour, is seen in projection on the *b*-plane. To a first approximation it can be regarded as a spheroid with a length of 64 Å., a width of 55 Å. and a height of 50 Å. normal to the plane of the paper. Except for a slight shortening along *a*, this shape agrees with the earlier picture obtained from two-dimensional data, even including the dimple in the centre of the molecule (see Fig. 2 of ref. 12).

Positions of the Hæm and Sulphydryl Groups

Four peaks stand out from the rest, clearly representing the iron atoms with their surrounding porphyrin rings. A flattening of the peaks indicates the approximate orientation of the rings (Fig. 3). Fortunately, their exact orientation was already known from electron spin resonance[14], and it only remained to assign the correct one of the four alternative orientations of the hæms to each of the electron density peaks. The results are shown in Fig. 4. The iron atoms lie at the corners of an irregular tetrahedron with distances of 33·4 and 36·0 Å. between symmetrically related pairs. The closest approach between symmetrically unrelated iron atoms is 25·2 Å. (Table 1).

Table 1

	Fe_1	Fe_3	S_1
x	−6·6 Å.	12·3 Å.	5 Å.
y	7·3 Å.	−10·7 Å.	10 Å.
z	13·1 Å.	18·2 Å.	16 Å.

$Fe_1 - Fe_2 = 33{\cdot}4$ Å.
$Fe_3 - Fe_4 = 36{\cdot}0$ Å.
$Fe_1 - Fe_3 = 25{\cdot}2$ Å.
$Fe_1 - S_1$ 13 Å.
$Fe_3 - S_1$ 21 Å.

Horse hæmoglobin contains four cysteine residues, but only two sulphydryls combine with mercury in the native protein[5]. From the positions of the mercury atoms we inferred that each of the two sulphydryl groups is about 13 Å. away from one iron atom and 21 Å. away from another. (Fig. 4 and Table 1). The significance of this situation is discussed below.

Configuration of the Polypeptide Chains

The most prominent feature of the Fourier synthesis consists of more or less cylindrical clouds of high density, like the vapour trails of an aeroplane; they are curved to form intricate three-dimensional figures. Sections through various parts of these appear in Fig. 1. To build a model of the figures, we rolled out sheets of a thermo-setting plastic to the thickness of our sections on a scale of 2 Å. = 1 cm., and cut out the shape of each region on the contour maps where the density exceeds 0·54 electron/Å.^{3} (this corresponds to the first full contour line in Fig. 1. From this section, for example, 14 shapes would be cut). The shapes were then assembled in accordance with their positions and heights in the different sections, and the hæm groups were attached in the appropriate orientation. The model was then baked to set it permanently. For comparison, a Fourier synthesis of sperm whale myoglobin was calculated at a resolution of 5·5 Å., using the new X-ray data of Kendrew *et al.*[15], and a model of the electron density distribution was constructed by the method just described.

From the hæmoglobin Fourier synthesis there emerged four separate units which are identical in pairs. Fig. 5 shows one member of each pair together with the model of myoglobin on the left. In each unit the cloud of high density describes a complicated figure which, in the white unit (middle), can be traced from end to end by following the superimposed line. Except for two small gaps where the density sinks slightly below 0·54 electron/Å.^{3}, the black unit (right) closely resembles the white one. There are several gaps interrupting the myoglobin model where, probably due to increased thermal motion, the electron density falls below the contour-level chosen for cutting the model. However, we know from the recent work of Kendrew and his collaborators[15] that

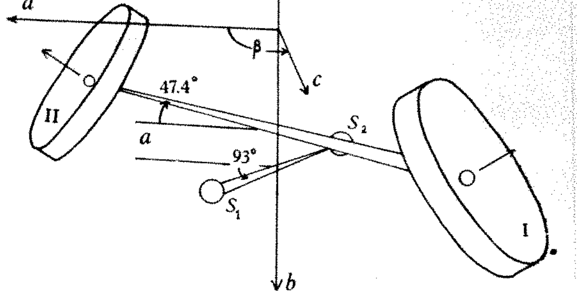


Fig. 4. Arrangement of hæm groups in hæmoglobin. Arrows indicate the reactive side of each group. The *c*-axis comes out of the paper towards the observer. This picture and all subsequent ones show the absolute configuration

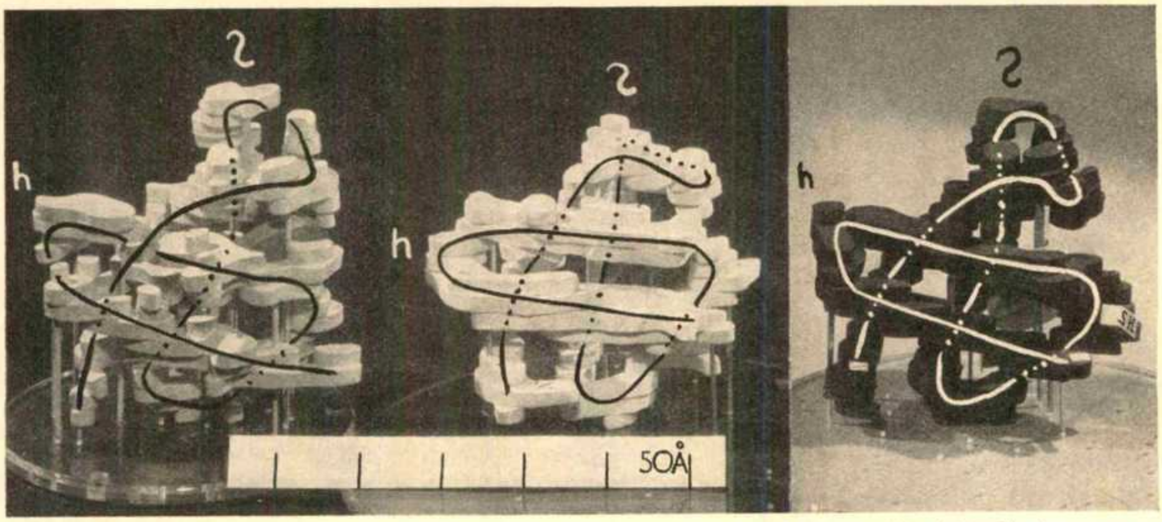


Fig. 5. Two different polypeptide chains in the asymmetric unit of hæmoglobin compared with myoglobin (left). The hæm groups are at the back of the chains

the gaps are bridged by a continuous polypeptide chain, and it is evident from Fig. 5 that apart from the gaps, the model has a configuration closely similar to the hæmoglobin units.

Clearly, the four tortuous clouds of high electron density in hæmoglobin represent the four polypeptide chains. The black and the white chains have similar, but not identical configurations. In the black chain the *S*-shaped bend at the top is more pronounced, the hæm group is lower, and the bend *h* sharper than in the white chain. These, however, are details. The most important result is their resemblance to each other and to sperm whale myoglobin.

Arrangement of the Four Sub-units

The first step in the assembly of the molecule is the matching of each chain by its symmetrically related partner (Fig. 6). It will be noted that there is comparatively little contact between the members of each pair, suggesting rather tenuous linkages. In the next step the white pair is inverted and

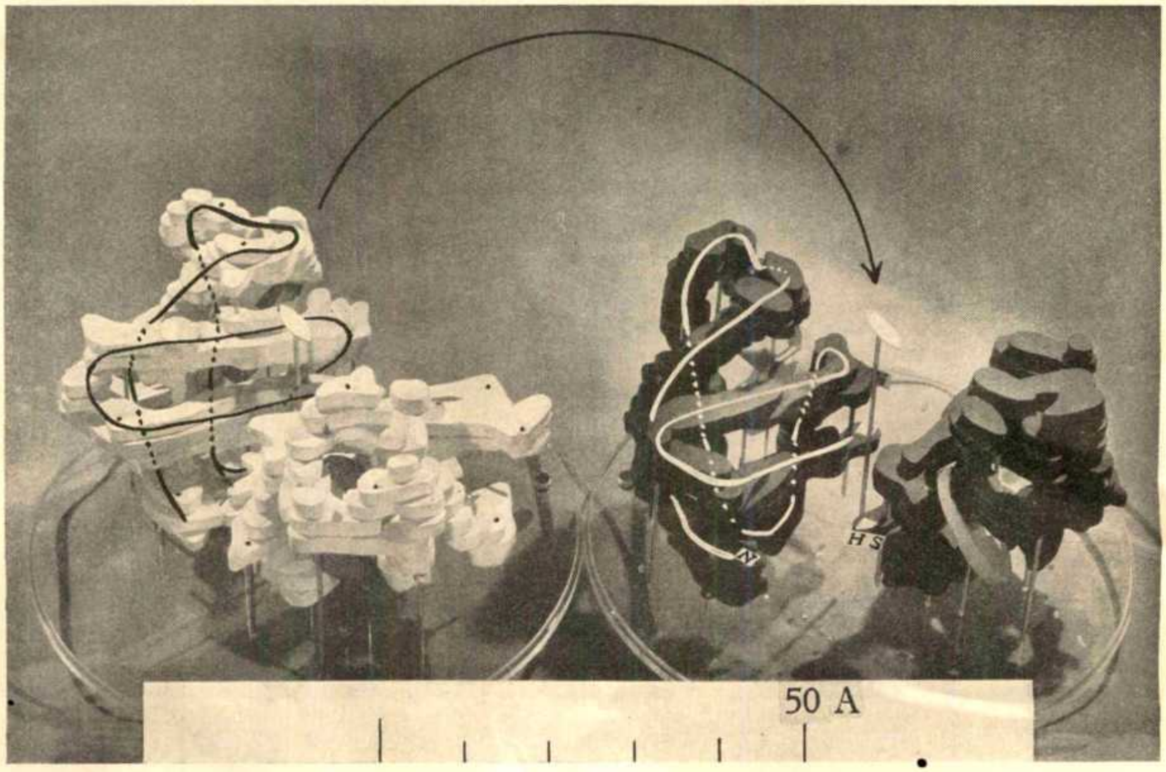


Fig. 6. Two pairs of chains symmetrically related by the dyad axis. The arrow shows how one pair is placed over the other to assemble the complete molecule

placed over the black pair as indicated by the arrow. Fig. 7 shows one white chain placed over the pair of black ones and Fig. 8 shows the molecule completely assembled. The resulting arrangement is tetrahedral and has almost, but not quite, the orthorhombic point group symmetry 222. It contains two 'pseudo dyads' which lie approximately at right angles to each other and to the true dyad, one emerging from the centre of Fig. 8 and the other from the centre of Fig. 9. This means that, to a first approximation, each sub-unit can be generated by a rotation of 180° from any of its neighbours. Figs. 7, 8 and 9 also show that the surface contours of the white chains exactly fit those of the black, so that there is a large area of contact between them. This structural complementarity is one of the most striking features of the molecule. Fig. 10 is a view down the true dyad axis and reveals a hole going right through the centre of the molecule, as was to be expected from the Fourier projection[13]. However, the van der Waals radii of the chains are much bigger than appears in the model, and little room may, in fact, be left for water or electrolytes to pass through. Fig. 10 also reveals a dimple at the top where the white chains meet. This is matched by a similar, but larger, hollow at the bottom where the black chains meet.

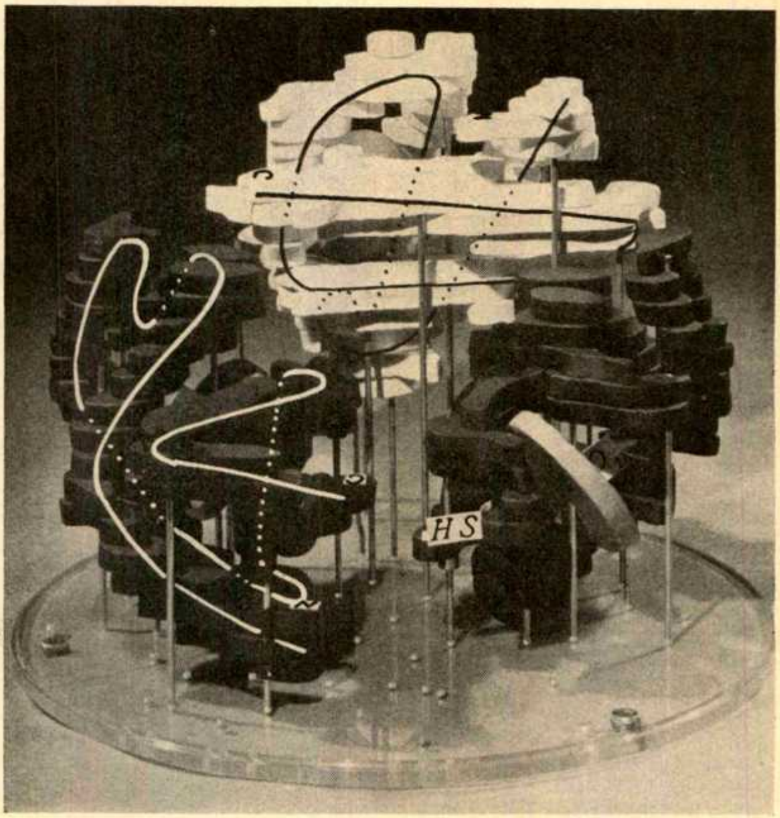


Fig. 7. Partially assembled molecule showing two black chains and one white

The hæm groups are seen to lie in separate pockets on the surface of the molecule (Fig. 8). Each pocket is formed by the folds in one of the polypeptide chains, which appears to make contact with the hæm group at four different points at least. The iron atoms in the neighbouring pockets formed by the black and the white chains are 25 Å. apart.

Information from the Fourier Synthesis of Myoglobin at 2-Å. Resolution (ref. 15)

Thanks to the similarity with myoglobin, the interpretation can be carried further than would have been possible on the basis of our results alone. Kendrew *et al.* have found that the straight stretches of rod indicated in Fig. 5 are α-helices and that the N-terminal end of the chain is at the bottom left. The ends of the two hæmoglobin chains have been labelled *N* and *C* accordingly, as seen in Figs. 7 and 10. The myoglobin Fourier synthesis also reveals the hæm-linked amino-acid side-chain, probably histidine, on one side of the (ferric) iron atom and a small peak, probably representing a water molecule, on the other side. If this information is transferred to hæmoglobin, the reactive side of the hæm group is as indicated by the arrows in Fig. 4 and by the labels O_2 in Figs. 7 and 8*a*.

Figs. 7 and 8*b* show the reactive sulphydryl group (which is absent in myoglobin) to be attached to the portion of the black chain carrying the hæm-linked histidine. It is seen that the histidine and cysteine side-chains point in roughly opposite directions, one towards and the other away from the hæm group. The sulphydryl group may possibly be in contact with the loop of the white chain which lies below the hæm group on the left. This is the situation of one pair of sulphydryl groups in the molecule. The second pair is probably attached to the white chains, but it is unreactive in the native protein and its position is still unknown.

Reliability of Results

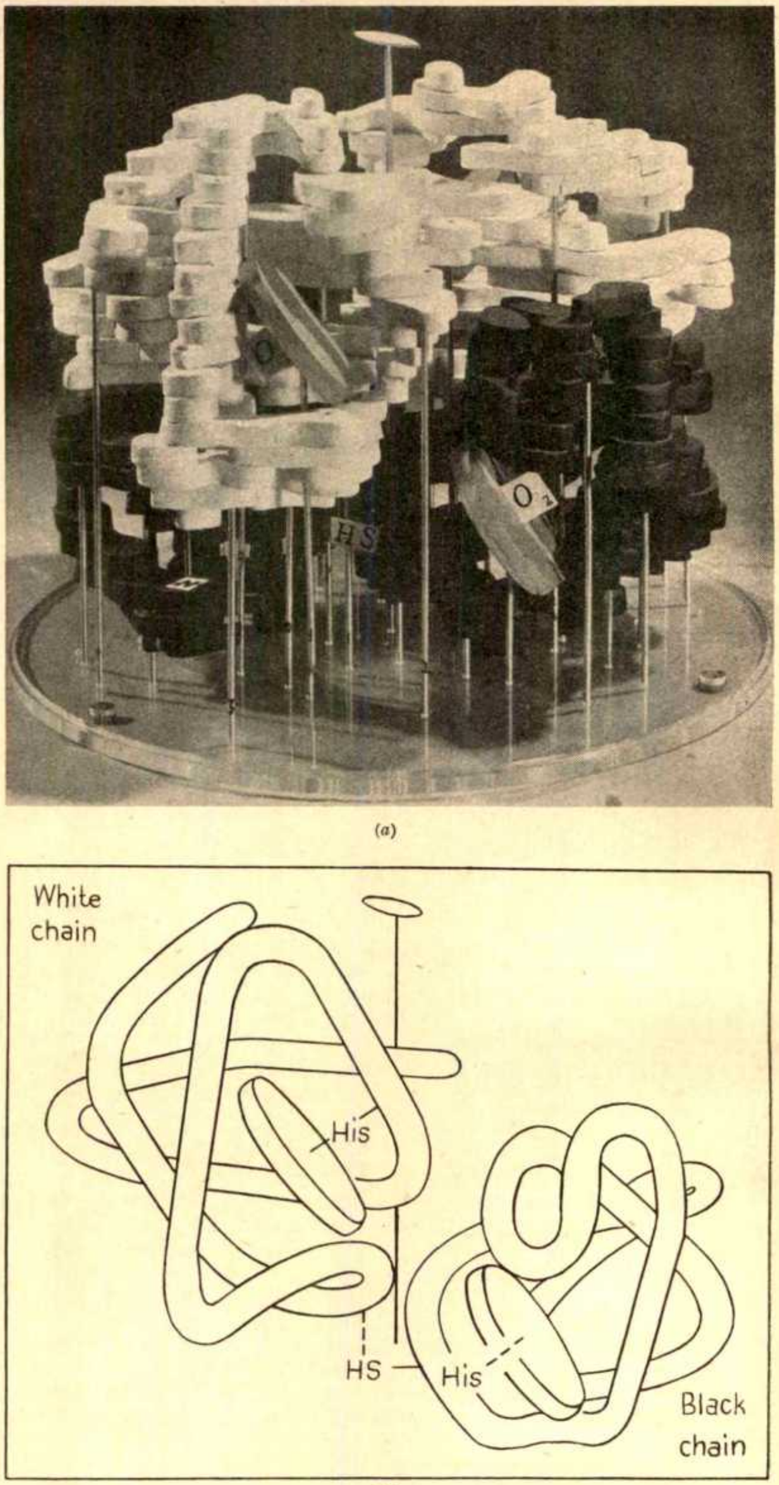
The method of isomorphous replacement makes no assumptions about the structure of the protein, and its results suffer from none of the ambiguities which bedevil the interpretation of vector maps. Ideally, the parent protein, in combination with two different isomorphous heavy-atom compounds, should give accurate phase angles and electron density maps which are free from all except series termination errors. In practice, errors and uncertainties arise from several sources and must be minimized by using more than two heavy-atom compounds. In our case it was possible to estimate the accuracy of the phase angle for each reflexion from the measure of agreement between the angles indicated by the six different compounds. From the standard error in the vector *F*, averaged over all reflexions, the standard error in the final electron density was calculated as $0\cdot12$ electron/Å.^{3}, which amounts to $0\cdot85$ of the interval between successive contours in Figs. 1 and 3 ($0\cdot14$ electron/Å.^{3}), or $0\cdot15$ of the difference in density between 'peaks' and 'valleys' ($0\cdot7$ electron/Å.^{3}) (ref. 12). This calculated error is borne out by the observed fluctuations in the liquid regions between the molecules and by the difference in height between the two iron peaks ($0\cdot13$ electron/Å.^{3}).

An error of at least five times this magnitude would be needed to turn a peak into a valley and thus to simulate the backbone of a polypeptide chain in a region actually occupied by side-chains.

Normally, the accuracy of a structure is checked by comparison of calculated and observed intensities. In hæmoglobin this is impossible so long as the light atoms are not resolved, but the positions of the iron atoms at least can be checked by their anomalous dispersion effect. Depending on the phase relationship between the structure factor of the four iron atoms by themselves and that of the entire molecule, the total intensity of any given reflexion $I\ (hkl)$ may be larger or smaller than $I\ (\bar{h}\bar{k}\bar{l})$. We selected 50 reflexions for which the effect of anomalous dispersion was expected to be largest and measured the intensity of each reflexion in the four symmetrically related quadrants on the counter spectrometer. Statistically significant differences between $I(hkl)+I(\bar{h}k\bar{l})$ and $I(\bar{h}\bar{k}\bar{l})+I(h\bar{k}l)$ were found in 36 reflexions. In 34 of them the signs of the differences agreed with prediction, indicating that the phase angles of the reflexions as well as the positions of the iron atoms are correct.

As a further check on the positions of the iron atoms, an attempt was made to label them with heavy atoms. This was done by allowing crystals of methæmoglobin, which are isomorphous with oxyhæmoglobin, to react with *p*-iodophenyl–hydroxylamine, which attaches itself to the iron atoms. A difference Fourier projection on the *b*-plane showed four prominent peaks which lie well within the calculated distances between the iodine and the iron atoms.

Finally, to avoid any possible bias in the interpretation of the Fourier synthesis, we have tried to construct objective models containing only those features actually found in the electron density maps. Due to series termination errors and inaccuracies in many of the phase angles, these maps must certainly contain errors of detail. Large errors, on the other hand, are unlikely because, quite apart from the checks just described, the results really prove themselves. No combination of errors could have led to the appearance of four distinct chains of roughly equal length, in agreement with chemical evidence, to the similarity between the two pairs of chains which are not related by crystal symmetry, and to the resemblance between hæmoglobin and myoglobin.



(*a*)

(*b*)

Fig. 8. (*a*) Hæmoglobin model viewed normal to *a*. The hæm groups are indicated by grey disks. (*b*) Chain configuration in the two sub-units facing the observer. The other two chains are produced by the operation of the dyad axis

Discussion

The polypeptide chain-fold which Kendrew and his collaborators first discovered in sperm whale myoglobin has since been found also in seal myoglobin[16].

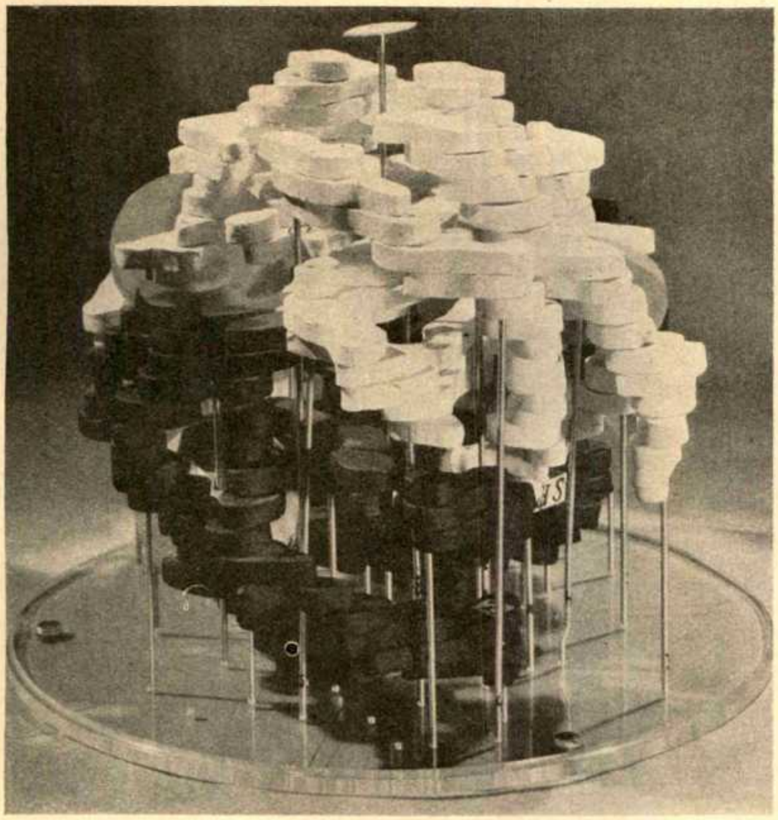


Fig. 9. Hæmoglobin model viewed normal to *c*

Its appearance in horse hæmoglobin suggests that all hæmoglobins and myoglobins of vertebrates follow the same pattern. How does this arise ? It is scarcely conceivable that a three-dimensional template forces the chain to take up this fold. More probably the chain, once it is synthesized and provided with a hæm group around which it can coil, takes up this configuration spontaneously, as the only one which satisfies the stereochemical requirements of its amino-acid sequence. This suggests the occurrence of similar sequences throughout this group of proteins, despite their marked differences in amino-acid content. This seems all the more likely, since their structural similarity suggests that they have developed from a common genetic precursor.

Little can be said as yet about the relation between structure and function. The hæm groups are much too far apart for the combination with oxygen of any one of them to affect the oxygen affinity of its neighbours directly. Whatever interaction between the hæm groups exists must be of a subtle and indirect kind that we cannot yet guess. A few observations of possible significance might be mentioned. Kendrew found that the combination of reduced myoglobin with oxygen involves no structural changes detectable by X-ray analysis[17], both forms being isomorphous with the metmyoglobin normally studied. In the hæmoglobin of horse and of man, on the other hand, the oxygenated and reduced forms are crystallographically different[18-20]. The structure of reduced hæmoglobin is still unknown, but it would not be surprising if loss of oxygen caused the four sub-units to rearrange themselves relative to each other, rather than to change their individual structure to a marked degree.

Riggs has shown that blocking the sulphydryl groups reduces hæm–hæm interaction[21]. As Fig. 8 shows, these groups occupy key positions close to the hæm-linked histidines and to points of contact between two different sub-units. They may well play an important part in the transition between the oxygenated and reduced forms. Incidentally, the cysteine residue should provide a convenient marker for the peptide containing the hæm-linked histidine, and so help to determine the sequence in this important part of the chain.

A full account of this work will be published elsewhere.

We thank the Director and staff of the University of Cambridge Mathematics Laboratory for making their electronic computor *Edsac* II available to us for the many calculations involved in this work. We are

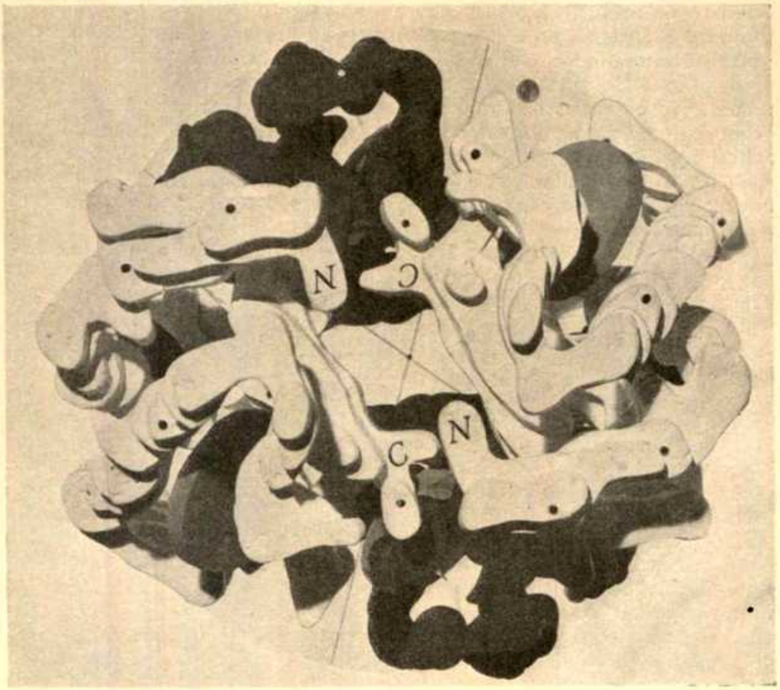


Fig. 10. A view down the *b*-axis. Note the proximity of the *C* and *N* terminal ends which could serve to form links between the two white chains

also deeply grateful to the Rockefeller Foundation for its long-continued financial support and to Sir Lawrence Bragg for his unfailing enthusiasm and encouragement. Finally, we wish to thank Dr. B. R. Baker, of the Stanford Research Institute, for a gift of organic mercurials, Dr. J. Chatt of Imperial Chemical Industries, Ltd., and Mr. A. R. Powell, of Johnson Matthey, Ltd., for gifts of heavy-atom compounds, and Mrs. Margaret Allen, Miss Ann Jury and Miss Brenda Davies for assistance.

[1] Rhinesmith, S. H., Schroeder, W. A., and Pauling, L., *J. Amer. Chem. Soc.*, **79**, 4682 (1957).

[2] Braunitzer, G., *Hoppe-Seyl. Z.*, **312**, 72 (1958).

[3] Wilson, S., and Smith, D. B., *Can. J. Biochem. Physiol.*, **37**, 405 (1959).

[4] Bokhoven, C., Schoone, J. C., and Bijvoet, J. B., *Acta Cryst.*, **4**, 275 (1951).

[5] Green, D. W., Ingram, V. M., and Perutz, M. F., *Proc. Roy. Soc.*, A, **225**, 287 (1954).

[6] Kendrew, J. C., Bodo, G., Dintzis, H. M., Parrish, R. G., Wyckoff, H. W., and Phillips, D. C., *Nature*, **181**, 662 (1958).

[7] Blow, D. M., *Proc. Roy. Soc.*, A, **247**, 302 (1958).

[8] Perutz, M. F., *Proc. Roy. Soc.*, A, **225**, 264 (1954).

[9] Cullis, A. F., Dintzis, H. M., and Perutz, M. F., Conference on Hæmoglobin, National Academy of Sciences, NAS-NRC Publication 557, p. 50 (Washington, 1958).

[10] Rossmann, M. G., *Acta Cryst.* (in the press).

[11] Bodo, G., Dintzis, H. M., Kendrew, J. C., and Wyckoff, H. W., *Proc. Roy. Soc.*, A, **253**, 70 (1959).

[12] Blow, D. M., and Crick, F. H. C., *Acta Cryst.*, **12**, 794 (1959).

[13] Bragg, W. L., and Perutz, M. F., *Proc. Roy. Soc.*, A, **225**, 315 (1954).

[14] Ingram, D. J. E., Gibson, J. F., and Perutz, M. F., *Nature*, **178**, 905 (1956).

[15] Kendrew, J. C., Dickerson, R. E., Strandberg, B., Hart, R. G., Davies, D. R., Phillips, D. C., and Shore, V. C. (see following article).

[16] Scouloudi, H., *Nature*, **183**, 374 (1959).

[17] Kendrew, J. C. (private communication).

[18] Haurowitz, F., *Hoppe-Seyl. Z.*, **254**, 266 (1938).

[19] Jope, H. M., and O'Brien, J. R. P., "Hæmoglobin", 269 (Butterworths, London, 1949).

[20] Perutz, M. F., Trotter, I. F., Howells, E. R., and Green, D. W., *Acta Cryst.*, **8**, 241 (1955).

[21] Riggs, A. F., *J. Gen. Physiol.*, **36**, 1 (1952).

STRUCTURE OF MYOGLOBIN

A THREE-DIMENSIONAL FOURIER SYNTHESIS AT 2 Å. RESOLUTION

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MYOGLOBIN is a conjugated protein consisting of a single polypeptide chain of about 153 amino-acid residues associated with an iron–porphyrin complex, the hæm group; its molecular weight is about 18,000, and the molecule contains some 1,200 atoms (excluding hydrogen). Two years ago a preliminary report of the first stage of an investigation of the three-dimensional structure of sperm-whale myoglobin was published in *Nature*[1] (a detailed account of this work has appeared recently[2]). Several isomorphous crystalline derivatives of myoglobin containing heavy atoms (mercury or gold) at single sites on the molecule were prepared, and by comparing the X-ray diffraction patterns of these crystals with those of the unsubstituted protein, it was possible to deduce the phases of all the reflexions in the X-ray pattern having spacings greater than 6 Å. These phases, together with the observed amplitudes, were used to compute a three-dimensional Fourier synthesis of the electron density in the unit cell (which contains two molecules) at a resolution of 6 Å. In this synthesis the polypeptide chain was visible as a rod of high electron density, folded in a complex pattern (Fig. 5*a*); in addition, the single hæm group with its iron atom, which is much more dense than any other atom in the molecule, could be identified as a disk of high electron density. The shape of the molecule could be determined with some confidence, as could most of the course of the single polypeptide chain, though there were several ambiguities where it turned through a large angle, so that the ends of the chain could not be located with certainty. Thus, the general nature of the tertiary structure of the molecule was revealed, but not the secondary structure of the polypeptide chain, though the results were consistent with a helical configuration.

More recently, Scouloudi[3] used similar methods to obtain a two-dimensional Fourier projection of the unit cell of seal myoglobin, in which the molecular arrangement is entirely different from that in sperm-whale crystals; she was able to show that the myoglobins of these two species have essentially the same tertiary structure in spite of their different amino-acid compositions. Her work, by implication, confirmed the correctness of both analyses, as well as the deductions made about the shape of the molecule. In the accompanying article[4] Perutz *et al.* now describe a three-dimensional analysis of the related protein hæmoglobin, at a slightly greater resolution, and show that each of the four sub-units of which this molecule is composed bears a close structural resemblance to myoglobin. It is apparent, therefore, that sperm-whale myoglobin possesses a structure the significance of which extends beyond a particular species and even beyond a particular protein.

We now present the results of a second stage in the analysis of sperm-whale myoglobin; in this the resolution has been increased to 2 Å., that is to say, not far short of atomic resolution. The resulting Fourier synthesis is very complicated, and a detailed study of it will take many months; in the meantime, our preliminary findings may be of interest.

Methods of X-ray Analysis

In this stage we have simply extended the methods which proved successful in the first stage of the analysis, comparing the diffraction pattern of unsubstituted myoglobin crystals with those of the *p*-chloromercuribenzene sulphonate, mercury diammine and aurichloride derivatives, together with a double derivative containing the first two substituents simultaneously. Whereas myoglobin crystals give 400 reflexions having spacings greater than 6 Å., the number of reflexions with spacings greater than 2 Å. is 9,600, each of which has to be measured not only for the unsubstituted protein but also for each of the derivatives. The very much greater number of data posed many problems, both in recording intensities and in computation, and in this stage we relied much more heavily than before on the use of a high-speed computer; it was fortunate that about the time the work began the *Edsac* Mark I computer used previously was superseded by the very much faster and more powerful Mark II.

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* Visiting scientist from the National Institute of Mental Health, Bethesda, Md.

The data for each derivative were recorded on twenty-two precession photographs; a separate crystal had to be used for each photograph to keep radiation damage within acceptable limits. The results from the different photographs were scaled together on the computer, the best set of scaling factors being determined by solving an appropriate 22 × 22 matrix[5]. The degree of isomorphism of each derivative was tested, and found adequate, by means of a computer programme which used the *h0l* reflexions to refine the preliminary values of the heavy-atom parameters, temperature factor, etc., and then compared the values of $\delta F_{\text{obs.}}$ and $\delta F_{\text{calc.}}$ as a function of $\sin\theta$. The co-ordinates of the heavy atoms were further refined using correlation functions[6] computed by means of programmes devised by Dr. M. G. Rossmann, and finally refined again during the process of phase determination itself. The phases were determined by essentially the same method as before, but owing to the very large number of reflexions the determination was carried out on the computer rather than graphically. The 'best' phases and amplitudes[7] were computed and used in the final Fourier synthesis, to which a moderate degree of sharpening was applied. In all, some hundreds of hours of computer time were required, and the Fourier synthesis itself, which was calculated at intervals of about 2/3 Å., took about 12 hr.

The Fourier Synthesis

The electron density distribution was plotted in the form of 96 sections perpendicular to x^* and spaced 2/3 Å. apart, the density in each section being represented by a series of contours. For some purposes this method of representation is unsatisfactory, and we have also constructed models of parts of the structure on a scale of 5 cm. = 1 Å., by erecting vertical steel rods parallel to y in an array corresponding to the grid of points in the xz-plane at which densities were calculated, the value of the electron density at points along the rods being indicated by coloured clips. On this scale the whole molecule is about 6 ft. cube, and about 2,500 rods each 6 ft. high are required (see Fig. 4).

The distance between singly bonded carbon atoms is 1·54 Å.; many of the common types of covalent bond present in protein molecules are shorter than this. We could not expect, therefore, that neighbouring covalently bonded atoms would be resolved in the present Fourier synthesis, and in fact such resolution was not achieved; on the other hand, the degree of resolution actually obtained is very nearly as great as theory would indicate, showing that the errors in the synthesis cannot be very large (see Fig. 2). The distances between neighbouring groups in Van der Waals contact is of the order of 3 Å., however, and as would be anticipated, we find that such groups appear as discrete regions of high density. One factor which complicates the interpretation is that similar groups (for example, peptide bonds) in different parts of the structure have substantially different densities. We attribute these differences to variation in the amplitude of thermal vibration in different parts of the molecule. Thus the terminal amino-end of the chain, which is on the outside of the molecule, has low density and is presumably relatively flexible; on the other hand, structures near the hæm group, which is surrounded by several segments of polypeptide chain running in different directions, possess high density, presumably because this part of the molecule is stabilized by many inter-chain bonds.

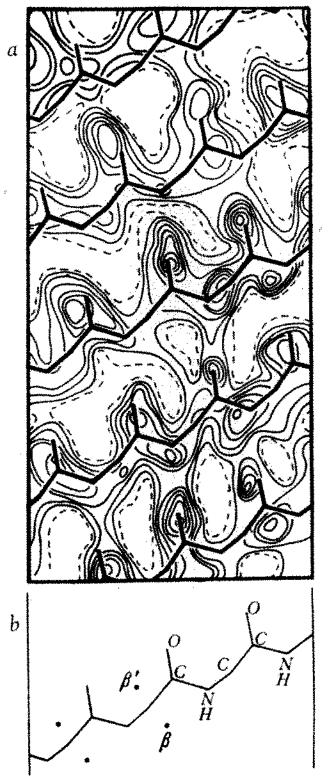

Fig. 1. (*a*) Cylindrical projection of a helical segment of polypeptide chain, with the α-helix structure superposed: for explanation see text. (*b*) Key to the atomic arrangement in the α-helix. The points marked β and β′ are the two alternative projected positions of C_β; β is the position in a right-handed and β′ that in a left-handed helix of L-amino-acids

Configuration of the Polypeptide Chain

In the 6-Å. Fourier synthesis the polypeptide chain appeared as a solid rod of high density with approximately circular cross-section; segments of the chain were more or less straight, and these were joined by dense regions where the chain turned corners. In the 2-Å. synthesis all the straight segments of chain were found to be hollow cylindrical tubes of high density; projecting at intervals from the cylindrical core are dense regions of various shapes and sizes, which are clearly the amino-acid side-chains. More detailed

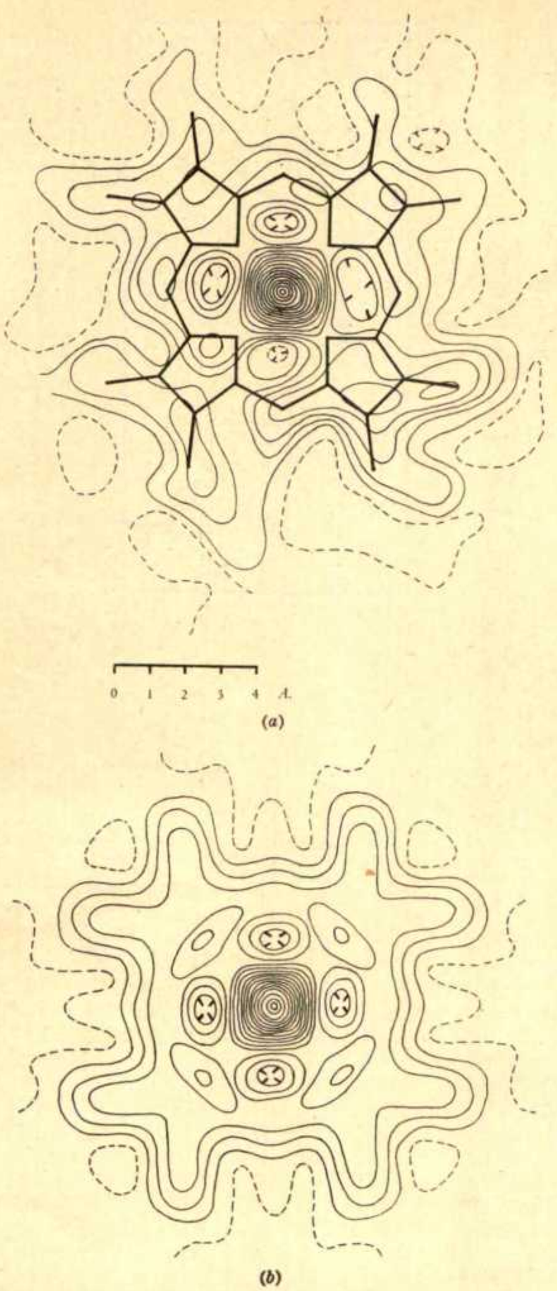

Fig. 2. (a) Observed electron density distribution in the plane of the hæm group, with the atomic arrangement in the group superposed. One contour interval = 0·5 electron/Å^3. (b) Calculated electron density distribution, computed from the known atomic arrangement as described in the text. One contour interval = 0·5 electron/Å^3

examination shows that, in fact, the cylindrical tubes are helices, consisting of a single strand of high density with axial repeat about 5·4 Å. The density in one such helix is shown in Fig. 1*a*. This was obtained by projecting all the density lying within a cylindrical shell of radii 1·6 and 2·3 Å. on to the surface of a cylinder of radius 1·95 Å. and co-axial with the helix; the cylinder was then cut along a line parallel to its axis and unrolled. Superposed on the contours of electron density is the cylindrical projection of an α-helix, with the dimensions given by Pauling and Corey[8]. It will be seen that the observed density follows the configuration of the α-helix with remarkable precision; and it may confidently be asserted that not only this, but also several other lengths of polypeptide chain which have been analysed in the same way, possess the α-helical configuration. It is also found that the side-chains emerge at intervals of 100° around the periphery of the helix, and at axial intervals of 1·5 Å., in conformity with the parameters of the α-helix. Several indirect lines of evidence had led to the conclusion that parts of the polypeptide chain in globular proteins are in the form of α-helixes, and in hæmoglobin Perutz[9] had found traces of an X-ray reflexion characteristic of this configuration; but these results are the first direct proof that α-helices are present and, indeed, enable them to be seen directly for the first time.

The next question is whether the α-helices are left-handed or right-handed; helices of either hand

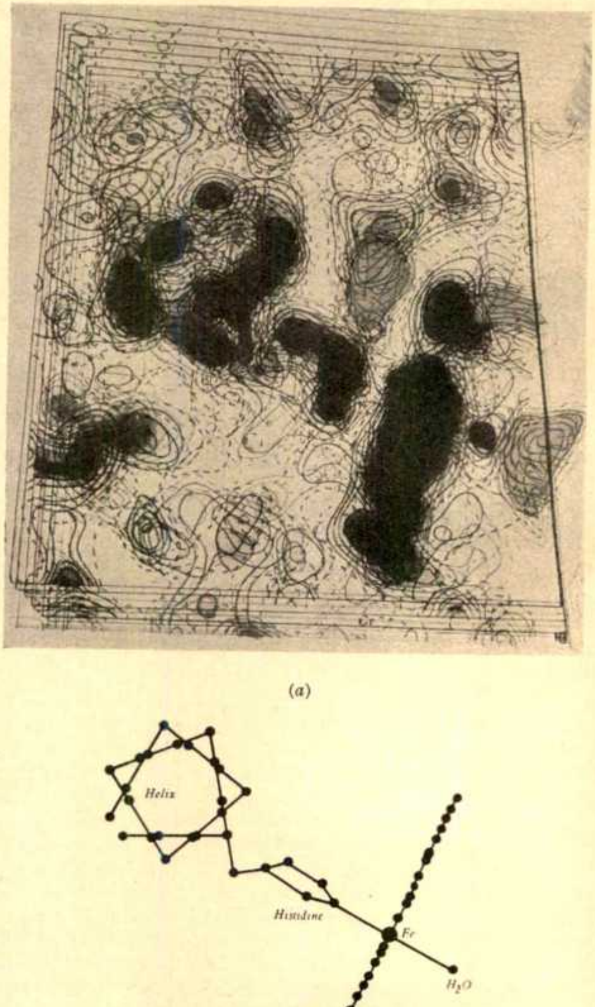

Fig. 3. (a) Photograph of a set of sections normal to the plane of the hæm groups showing, from left to right, a helix in cross-section, the histidine residue nearly edge-on, the hæm group edge-on, and a presumed water molecule. (b) Sketch showing the atomic arrangement in Fig. 3*a*

can be built, and although the right-handed form appears to be marginally the more stable, the difference is not so great as to make it certain that left-handed helices cannot exist. To decide this question, it is necessary to determine the absolute configuration of the Fourier synthesis; this function has no centre or plane of symmetry and might be plotted in two ways, each of which would be the mirror image of the other. The absolute configuration could be settled by making use of the anomalous dispersion of X-rays from the iron atoms or the introduced heavy atoms; we have not in fact made use of this effect, but instead have proceeded in the following way. In a right-handed α-helix composed of L-amino-acids, the first atom of the side-chain (C_β) projects from the main chain in a direction *opposite* to the C—O bonds of the carbonyl groups; in a left-handed helix of L-amino-acids C_β projects in the *same* direction as the carbonyl groups (see Fig. 1*b*). By constructing a second cylindrical projection (not illustrated) of the density near a radius of 3·34 Å., corresponding approximately to the radius at which C_β is found in either case, we found that C_β was systematically on the side of the main chain opposite the oxygen atoms of the carbonyl groups. This shows that the helix must be right-handed, and we were able to plot the Fourier synthesis with the correct absolute configuration by taking account of the known absolute configuration of a L-amino-acid[10]. The molecule was then found to be of the same hand as all the closely similar sub-units in hæmoglobin, the absolute configuration of which was determined by measurements of anomalous dispersion[4]. All the lengths of α-helix in the myoglobin molecule turn out to be right-handed. Finally, it will be clear from Fig. 1 that it is possible to determine by inspection the direction in which the C—O group points, and hence to see which is the terminal carboxyl end of each segment.

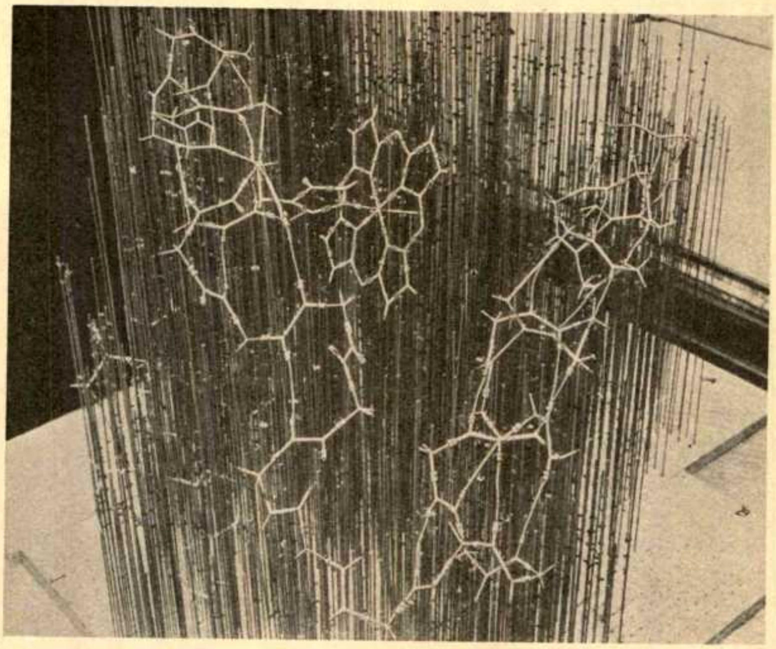

Fig. 4. Photograph of a model of part of the molecule near the hæm group, showing the vertical rods and coloured clips which indicate the electron density at each point of the grid, and atomic models of the hæm group and neighbouring helices. The terminal carboxyl end of the chain is on the extreme left

The α-helices can also be located by building atomic models into the model of the unit cell made with steel rods; this has been done for all the segments of helix in the molecule, and it is found that the total number of amino-acid residues contained in these segments is 100–110, whereas the number of residues contained in the whole molecule is believed to be 153 (Edmundson, A., unpublished results). Thus, 65–72 per cent of the molecule consists of regular right-handed α-helix, made up of about eight segments, each containing between seven and about twenty residues.

When the chain turns a corner, its regular helical configuration is necessarily disrupted. At the present resolution the precise arrangement of the residues at corners is difficult to determine; we have, nevertheless, built plausible models of several corners, though we do not yet claim to have established their configurations with certainty. Most of the corners take up two or three residues, and in addition there is one region (on the extreme right of Fig. 5*a*), consisting of about 13–18 residues, in which the arrangement is irregular (part of it is helical, but probably not α-helical). Further studies will be devoted to elucidating these problems.

No serious attempt has yet been made to identify the side-chains, and indeed it is doubtful whether this can be done systematically at the present resolution, partly because many of them appear to be subject to considerable thermal vibration, partly because there are often interactions between two side-chains on adjacent turns of the same helix or on adjacent helices, and it is difficult to tell where one ends and the other begins. In special cases, however, identification is easy; thus, in several places two helices approach one another closely, and one or more side-chains must for steric reasons be glycine. On the other hand, some side-chains are so long that they can only be arginine or lysine. It remains to be seen how many positive identifications can be made; but it is clear that if the whole amino-acid sequence of the protein were known, it would be possible at this stage to construct a model of the complete structure with fair precision. Unfortunately, this information is not yet available; but in the meantime it may be possible to correlate and check our findings with the partial results of the sequence determination now being undertaken by Mr. A. Edmundson, of the Rockefeller Institute.

The Hæm Group

The identification of a disk of high electron density in the 6-Å. synthesis as the hæm group is confirmed by the 2-Å. synthesis; but the iron atom is now just resolved from the nitrogens of the porphyrin ring (the iron–nitrogen distance is about 1·9 Å.) and the structure of the group as a whole corresponds closely with theoretical expectation. Fig. 2*a* shows a section through the density distribution, cut in the plane of the hæm group; superposed on the section is a model of the hæm group with the dimensions

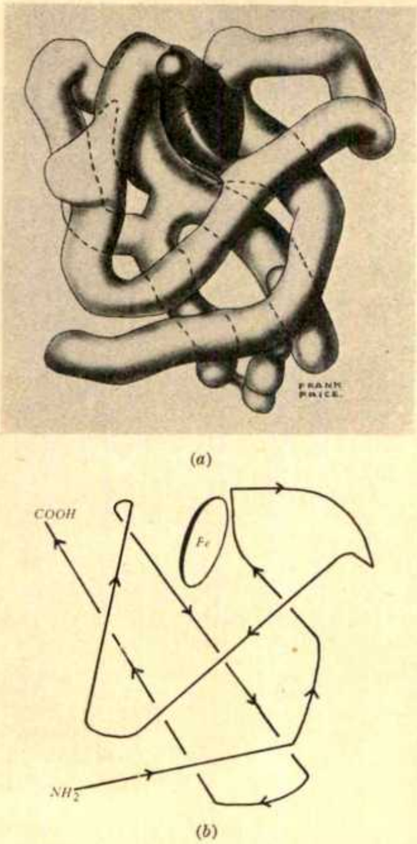

Fig. 5. (a) Drawing of the tertiary structure of myoglobin as deduced from the 6-Å. Fourier synthesis. (Note: the plane of the hæm group is indicated incorrectly in this drawing—see text). (b) The course of the polypeptide chain, deduced from the 2-Å. Fourier synthesis

found by Crute in his recent structure analysis of nickel etioporphyrin[11]. In the model, only the carbon atoms of the methyl groups and the first carbon atoms of the vinyl and propionic acid groups have been indicated; our analysis has not yet proceeded far enough to identify the latter groups. Fig. 2b is a Fourier synthesis made by calculating structure factors for the model structure in a convenient unit cell, applying to these the same temperature factor as that observed in myoglobin, and using them as terms of a Fourier synthesis cut off at 2-Å. spacings. It will be seen that the calculated and observed density distributions agree very closely; furthermore, the observed peak density (8·8 electrons/Å.^{3}) and half-width (1·8 Å.) of the iron atom agree well with the predicted values (8·2 electrons/Å.^{3} peak density, 1·8 Å. half-width). Errors are, of course, inherent in a Fourier synthesis produced from experimental results; the good agreement between the observed and calculated versions of the hæm group, and between the helical polypeptide chain and the α-helix (Fig. 1), indicate that in the present case these errors are fairly small, certainly less than we had anticipated.

In one respect the 6-Å. model now requires correction. Measurements of electron spin resonance[12] had shown that the two hæm groups in the unit cell were tilted out of the *bc* plane through 21° about an axis parallel to *c*, one in one direction and one in the other, but from these measurements one could not decide which direction of tilt was associated with the hæm group of which molecule. In the 6-Å. analysis the wrong choice was made; the new results show that the angle of tilt is indeed very close to 21°, but that it should be in the direction opposite to that shown in earlier models (including Fig. 5a).

Chemical studies of hæmoglobin and myoglobin have suggested that the hæm group is attached to the protein by a bond from the iron atom to a nitrogen atom of a histidine residue, and that the sixth co-ordination position of the iron atom is occupied in met-myoglobin (at *p*H's below 7) by a water molecule. Our results are in conformity with these suggestions. Fig. 3a is a photograph of a series of sections cut through the hæm group normal to its plane. The hæm group is seen edge-on; to its left is the histidine residue, also nearly edge-on, and to the left of that is the helix to which the histidine is attached, seen in cross-section. To the right of the hæm group is a small isolated peak which we take to be the oxygen atom of the water molecule (see Fig. 3b). When this structure is built with atomic models (see Fig. 4), it is found to satisfy all known requirements as to bond-lengths, angles, etc. Thus the iron–oxygen distance is close to 2·1 Å.; that between the iron and one of the histidine nitrogen atoms is 1·9 Å., while the second histidine nitrogen points with its hydrogen directly at a carbonyl group on the helix, presumably forming a hydrogen bond (forked at the carbonyl end); and finally the bonds about the C_α—C_β bond of histidine are almost exactly in the staggered configuration.

General Configuration of the Molecule

In the 2-Å. synthesis the continuity of the polypeptide chain can be followed throughout the molecule; in addition, its direction can be ascertained in each straight segment by examining the direction of the carbonyl groups in the cylindrical projections or by direct model-building. The 'run' of the chain so deduced everywhere agrees with that derived from the 6-Å. synthesis, as well as resolving several ambiguities in that model, and the terminal amino- and carboxyl-ends of the chain can now be identified. The arrangement is shown in Fig. 5 (see also Fig. 5 in the accompanying article by Perutz *et al.*). The overall shape of the molecule is as previously deduced.

Conservative estimates suggest that it may now be possible to locate with some precision at least 43 per cent of all the atoms (excluding hydrogens) in the molecule—namely, those making up the hæm group and the backbone of the regular segments of helix, together with the associated C_β's. We believe that it may be possible, either by straightforward refinement procedures or by heavy-atom methods, to extend the resolution of the synthesis to a point near the limit of the diffraction pattern of sperm-whale myoglobin crystals (about 1·5 Å.), and thus to reveal in any event most of the details of the atomic arrangement of the molecule. Further work is in hand with this end in view.

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[1] Kendrew, J. C., Bodo, G., Dintzis, H. M., Parrish, R. G., Wyckoff, H. W., and Phillips, D. C., *Nature*, 181, 662 (1958).

[2] Bodo, G., Dintzis, H. M., Kendrew, J. C., and Wyckoff, H. W., *Proc. Roy. Soc.*, A, 253, 70 (1959).

[3] Scouloudi, H., *Nature*, 183, 374 (1959).

[4] Perutz, M. F., Rossmann, M. G., Cullis, A. F., Muirhead, H., Will, G., and North, A. C. T. (see preceding article).

[5] Dickerson, R. E., *Acta Cryst.*, 12, 610 (1959).

[6] Rossmann, M. G., *Acta Cryst.* (in the press).

[7] Blow, D. M., and Crick, F. H. C., *Acta Cryst.*, 12, 794 (1959).

[8] Pauling, L., and Corey, R. B., *Proc. U.S. Nat. Acad. Sci.*, 37, 235 (1951).

[9] Perutz, M. F., *Nature*, 167, 1053 (1951).

[10] Trommel, J., and Bijvoet, J. M., *Acta Cryst.*, 7, 703 (1954).

[11] Crute, M. B., *Acta Cryst.*, 12, 24 (1959).

[12] Ingram, D. J. E., and Kendrew, J. C., *Nature*, 178, 905 (1956).

DEVELOPMENTS IN GAMMA-RAY OPTICS

By Prof. P. B. MOON, F.R.S.

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DURING the past decade, several phenomena that had previously been observed with light or with X-rays have been studied with nuclear γ-rays, where the quantum energy is typically 100 keV.–10 MeV. and the wave-length correspondingly short, 10^{-9} to 10^{-11} cm. Some new phenomena, specifically nuclear or specifically high-energy, have been added: but most of the experiments, though often technically more difficult, are extensions of those familiar at lower energies.

An excellent example of such an extension is the curved-crystal γ-ray spectroscopy of γ-rays, achieved with great elegance and precision by DuMond[1] and his collaborators. It will suffice to quote their value for the wave-length of electron–positron annihilation radiation, namely, 24·262 ± 0·0033 mÅ., and to mention that this radiation, when generated within a metal, has been observed to possess a spectral width which reflects the spread of energy of the conduction electrons.

A second example is the observation in the γ-ray region of Rayleigh scattering, a name taken over from visual optics to describe scattering of photons by the electronic structure of an atom without change of the atom's quantum state and therefore without any change of the photon's energy, except the small fraction taken by the recoil of the atom necessary to conserve momentum. This is the 'unmodified line' of X-ray scattering terminology, as opposed to the 'modified line' that arises from Compton scattering. The existence and (if it existed) the origin of elastic scattering at higher energies was a matter of controversy in 1930–33, but it is now clear that Meitner and Kösters observed it[2] and that its origin was electronic, not nuclear. In taking up the study of this scattering process with the wider range of mono-energetic lines provided by artificial radionuclides, Moon[3], Storruste[4] and Wilson[5] were able to verify semi-quantitatively the calculations of Franz[6] regarding its intensity and angular distribution, and many other workers have since made accurate experiments at energies above 1 MeV. where Franz's theory (which is essentially a Debye form factor) must be replaced by the more accurate relativistic treatment of Brown, Peierls and Woodward[7].

Two phenomena not observable in the X-ray region were predicted to be interwoven with the Rayleigh scattering; first, the classically calculable 'Thomson' scattering of photons by the nucleus, which oscillates in the electric field of the incident wave and radiates at the same frequency; secondly, scattering by virtual pair production in the field of the nucleus, which is a specifically quantum phenomenon.

The interest in the nuclear Thomson scattering seems to date from 1950, when it was remarked[8] that it should be coherent with Rayleigh scattering, but the process must surely have been considered long before then; the virtual pair production process was suggested as long ago as 1933 by Delbrück in a note to the paper of Meitner and Kosters[2].

Both processes were difficult to identify because the scattering radiation is of exactly the same frequency as the Rayleigh scattering but, at ordinary γ-ray energies, of much smaller intensity. The Thomson scattering was plausibly identified by the fact that the observed angular distribution of the total elastic scattering agreed much better with theory when this component was added into the calculation; its existence and its interference with the Rayleigh component were positively established by Sood's[8] measurements of the polarization of the elastic scattering at about 1 MeV., at which energy the Rayleigh and nuclear Thomson components have substantially different polarizations. The use of polarization for examining elastic scattering of γ-rays is pleasingly reminiscent of Lord Rayleigh and the blue of the sky.

The characteristic features of the Delbrück process are that its magnitude increases as a high power of the quantum energy and its angular distribution is strongly peaked around 0°. It has at last been unquestionably observed by Moffatt and Stringfellow[9] in Oxford, using 90-MeV. radiation from an electron synchrotron and working at angles of about 0·1°.

Another process of elastic scattering of γ-rays is that associated with the excitation of a nucleus from its ground-state to an excited state followed by the re-emission of a photon. This is the nuclear analogue of the optical resonance radiation so beautifully demonstrated by R. W. Wood in the early years of this century. It was sought experimentally at intervals over twenty years, the difficulty being not that the process is intrinsically weak (its cross-section, like that of any comparable resonant process, can be of the order of $\lambda^2/4\pi$) but that γ-ray lines are so narrow that the loss of energy to nuclear recoil ($h^2\nu^2/2Mc^2$ on emission and equal amount on absorption) destroys the resonance[10].

It was observed ten years ago[10] by the use of a high-speed rotor (not an ultracentrifuge, as it is called by some writers; the correct term is probably 'motor', since it is motion, not specifically rotary motion, and certainly not centrifugal force, that is needed) on which the source of γ-rays was mounted, so that the Doppler effect of bulk motion of the sources brought the emission spectrum into coincidence with the resonance of the scatterer. Both the emission and the absorption lines are broadened far beyond their natural width by the Doppler effect of thermal motion. Experimental results obtained by Davey and Moon[11] demonstrated the essentially thermal shape of the mercury-198 resonance line; its width is less than 1 eV. in 411 keV., which is comparable with the fractional width of the narrowest visible lines.

The phenomenon has been observed with several means of securing partial overlap of absorption and emission lines: by heating the source[12], by using atomic recoil from a preceding beta, gamma or neutrino emission[13–15] or by absorption of the resonance radiation in a second layer of the same material[14]. It has proved valuable for measuring the life-times of nuclear levels in the range 10^{-10} to 10^{-14} sec., and details such as the angular distribution[16] and the plane polarization[17] of the resonance radiation at 90° have been duly verified.

When the excited state decays largely through an intermediate level, the fluorescent radiation can be observed[15]. A particularly interesting application of the neutrino-recoil method was made by Goldhaber, Grodzins and Sunyar[22], who showed that in special circumstances the resonance radiation can be circularly polarized; they used this fact to determine the spin polarization (helicity) of the neutrino.

One is not restricted to γ-rays from radioactive sources: nuclear reactions, with the forward momentum of the excited nucleus as the overlapping agent, have been used both in scattering[18] and in absorption[19,20] experiments, and bremsstrahlung has been used to excite a particularly broad level[21].

A major development was reported in 1958 by Mössbauer in Munich[23]. He showed, using the 121-keV. line of iridium-191, that in crystals at low temperatures appreciable fractions of the emission and resonant absorption processes for low-energy γ-rays occur without nuclear recoil and indeed with no loss of energy between the γ-ray and the crystal other than that due to the recoil of the crystal as a whole; for these fractions, the line has only its natural width and may be brought out of resonance by relative motion of source and absorber of the order of 1 cm./sec. as compared with the order of 10^4 cm./sec. required to sweep the thermally broadened components over one another. The Mössbauer line sits as an extraordinarily tall and narrow spike on top of the thermal profile, at the same position for both emission and absorption. The cross-section for the Mössbauer effect is a sizable fraction of its ideal value ($\sim \lambda^2/4\pi$) and, even at 100 keV., can be appreciable in comparison not merely with the Rayleigh but also with the Compton and photoelectric cross-sections.

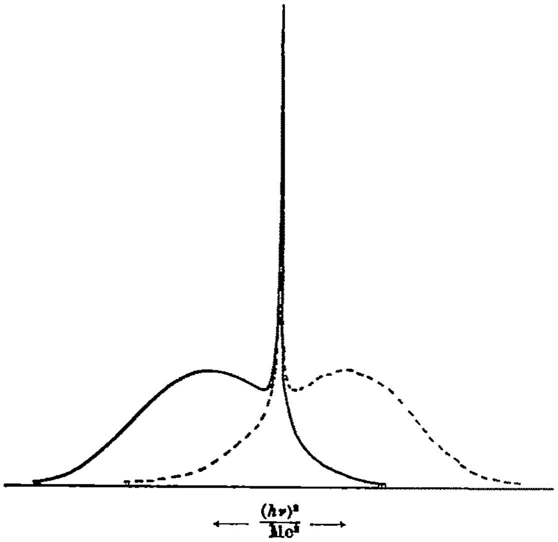

Fig. 1. Emission line profile (full line) and absorption line profile (broken line). The narrow line is present only with low γ-ray energies and cold crystals, when it may be $\sim 10^6$ times as high as the surrounding thermally broadened line and contain a large fraction of the total area

It is a curious reflexion on the journal-reading habits of physicists that Mössbauer's discovery was not generally known for a year after its publication; during the past few months, workers in many laboratories have realized its exciting possibilities. These are of two classes: its use to investigate the response of crystals to the offer of amounts of energy comparable with $k\theta$, where k is Boltzmann's constant and θ the Debye temperature, and the much more varied class of experiments that exploit the extreme narrowness of the line.

The effect has recently been observed in several laboratories with the 14 keV. line in iron-57, which has a width of the order of 10^{-8} eV.—one part in 10^{12} of its energy. It is, in fact, extremely easy to demonstrate, requiring only a thin source of cyclotron-irradiated iron, an absorber of ordinary iron, and a proportional counter or scintillation counter. Even at room temperature, changes of counting rate of the order of 30 per cent are observed when the source is oscillated at a speed of less than a millimetre per sec.

Perhaps the most exciting application of the Mössbauer effect is to observe the effect of gravitational fall on the energy of the photons (the 'terrestrial red-shift experiment') which appears measurable over a vertical distance of less than 100 m.: such experiments are understood to be in progress in the United States, England (Harwell and Manchester)[24] and the U.S.S.R. Another interesting experiment, being tried at Harwell and Birmingham, relates to special rather than general relativity; the source is mounted at the centre of a spinning rotor, the resonant absorber at the edge so that, though the distance between them does not change, the photons meet an absorber travelling exactly at right angles to their direction of flight: a second-order Doppler effect ($\frac{1}{2}v^2/c^2$) should be observable.

Mössbauer lines will have structures attributable to the interaction of the crystal's intrinsic magnetic or electric field with the magnetic dipole or electric quadrupole moments of the ground and excited states of the nucleus; Pound and Rebka[24] have already observed the splitting of the iron-57 line in the intrinsic magnetic field of iron. This is distinct from, and much narrower than, the structures due to the excitation of phonons in the crystal which, as Mössbauer pointed out, are analogous to effects observed in the scattering of very slow neutrons in crystals. In favourable instances the nuclear Zeeman effect should be observable.

One may imagine, too, searches for effects of electric or magnetic fields upon any small electric or magnetic moments that may be possessed by free photons, and one can see interesting problems concerning the limitations to the possible narrowness of Mossbauer lines from transitions still longer-lived than the 10^{-7} sec. iron-57. Are these limits due to fluctuations in crystal fields, to the finite size of the crystal (or of whatever part of it may act as a unit in declining to accept the proferred recoil energy) or to other causes? If the relative motion of the source and the absorber is at a frequency high compared with the inverse lifetime of the excited state, will the process recognize or ignore this motion?

The recoilless absorption process should be accompanied by sharply resonant scattering, weakened by the strong internal conversion of low-energy gamma transitions. In experiments at Birmingham, with gold-197, it appears just observable. This scattering should be coherent with the Rayleigh component and, in favourable circumstances, should show the lattice interference patterns familiar in X-ray work from which not only the positions of the resonantly scattering nuclei but also the phase of scattering should be measurable.

Thus, after a long period of little progress, γ-ray optics now offers a range of phenomena comparable with those known in the fields of light and of X-rays, and, though admittedly in rather special circumstances, a far superior resolving power.

[1] DuMond, J. W. M., in "Beta- and Gamma-Ray Spectroscopy", ed. Siegbahn (North Holland Pub. Co., 1955).
[2] Meitner, L., and Kösters, H., *Z. Phys.*, **84**, 137 (1933).
[3] Moon, P. B., *Proc. Phys. Soc.*, A, **63**, 1189 (1950).
[4] Storruste, A., *Proc. Phys. Soc.*, A, **63**, 1197 (1950).
[5] Wilson, R. R., *Phys. Rev.*, **90**, 721 (1953).
[6] Franz, W., *Z. Phys.*, **98**, 314 (1935).
[7] Brown, G. E., Peierls, R. E., and Woodward, J. B., *Proc. Phys. Soc.*, A, **227**, 51 (1954).
[8] Sood, B. S., *Proc. Roy. Soc.*, A, **247**, 375 (1958).
[9] Moffatt, J., and Stringfellow, M. W., *Phil. Mag.*, **3**, 540 (1958).
[10] Moon, P. B., *Proc. Phys. Soc.*, A, **64**, 76 (1951).
[11] Davey, W. G., and Moon, P. B., *Proc. Phys. Soc.*, A, **66**, 956 (1953).
[12] Malmfors, K. G., *Ark. Fysik*, **6**, 49 (1952).
[13] Moon, P. B., and Ilakovac, K., *Phys. Rev.*, **93**, 254 (1954). Ilakovac, K., *Proc. Phys. Soc.*, **67**, 601 (1954). Metzger, F. R., and Todd, W. B., *Phys. Rev.*, **95**, 853 (1954).
[14] Metzger, F. R., *Phys. Rev.*, **103**, 968 (1956).
[15] Grodzins, L., *Phys. Rev.*, **109**, 1014 (1958).
[16] Metzger, F. R., *Phys. Rev.*, **97**, 1258 (1955).
[17] Knapp, V., and Sood, B. S., *Proc. Roy. Soc.*, A, **247**, 369 (1958).
[18] Swann, C. P., and Metzger, F. R., *Phys. Rev*, **108**, 982 (1957).
[19] Hanna, S. S., and Meyer-Schutzmeister, L., *Phys. Rev.*, **108**, 1644 (1957).
[20] Smith, P. B., and Endt, P. M., *Phys. Rev.*, **110**, 1224 (1958).
[21] Hayward, E., and Fuller, E. G., *Phys. Rev.*, **106**, 991 (1957).
[22] Goldhaber, M., Grodzins, L., and Sunyar, A. W., *Phys. Rev.*, **109**, 1015 (1958).
[23] Mössbauer, R. L., *Z. Phys.*, **151**, 124 (1958); *Naturwiss.*, **45**, 538 (1958); *Z. Naturforsch.*, **14a**, 211 (1959).
[24] Pound, R. V., and Rebka, G. A., *Phys. Rev. Letters*, **3**, 554 (1959). Schiffer, J. P., and Marshall, W., *ibid.*, **3**, 556 (1959).
[25] Kuhn, W., *Phil. Mag.*, **8**, 625 (1929).

OBITUARIES

Prof. P. Lebeau

PAUL LEBEAU, professor of pharmaceutical chemistry, Paris, and member of the Paris Academy of Sciences, died on November 18, having nearly reached the age of ninety-one.

Lebeau began his career in chemistry as a student at the École de Physique et de Chimie, Paris, and his outstanding qualities were immediately appreciated by Prof. Étard. His early publications attracted the attention of H. Moissan, and when the latter was appointed to the chair of chemistry in the Faculté des Sciences at Paris, Lebeau was made director of his laboratory. His researches in inorganic chemistry, particularly with regard to fluorspar and fluorine, led to the degree of D.Sc. in 1898. During this period, he was also studying to become a pharmacist, and he achieved this object in 1899.

Lebeau was appointed to the chair of toxicology in 1908, which position he held until 1918. He deliberately orientated this subject towards industrial hygiene and became intensely interested in its medico-legal aspects. As a result, his services during the First World War were of outstanding merit, and in 1918 he succeeded Prof. Charles Moureu in the Berthelot chair in the Collège de France. His great energy and outstanding ability were then directed towards the subject of pharmaceutical chemistry, and his efforts met with similar international recognition as they had already done in the realm of toxicology. His treatise on pharmaceutical chemistry has passed through many editions and is accepted as a standard work on this subject.

As president of the Commission des Hautes Températures of the Centre National de la Recherche Scientifique he was largely responsible for the outstanding success of this body, which resulted in the publication in 1924 of an important book entitled "Fours Électriques et Chimie". This work has attained international recognition.

Not unnaturally, the services of a scientist of such outstanding merit were in great demand, particularly in connexion with government and international committees. These services were given without restraint, and Lebeau made outstanding contributions during his connexion with the Commission des Études Chimique de Guerre, the Conseil d'Administration du Centre National de la Recherche Scientifique, the Commission Internationale des Poids Atomique, and the Société Chimique de France.

Prof. Lebeau's long life covered the period when the science of chemistry was expanding rapidly, a period which was characterized by the introduction of many new chemical concepts. He played his part in this new thinking and can be regarded as one of the architects of modern pharmaceutical chemistry. His career and devotion to the science should be a stimulant to all future students of chemistry.

W. H. LINNELL

Mr. P. R. Coursey

MR. PHILIP RAY COURSEY, of the Dubilier Condenser Co., Ltd., died on January 3. Mr. Coursey, who was born in 1892, was educated at University College, London, and became assistant to Sir Ambrose Fleming at the College. During the First World War he was Admiralty inspector of wireless telegraphy in H.M. Auxiliary Patrol, and in 1919 became technical research assistant at H.M. Signals School. During

1920–23 he was on the editorial staff of the *Radio Review* and the *Wireless World*, to the second of which journals he was a frequent contributor. During this period he was an active and enthusiastic member of the Wireless Society of London (later, the Radio Society of Great Britain); and he was responsible for organizing the British amateurs who in 1921 co-operated in the tests conducted by the American Radio Relay League for establishing radio communication between the United States and Britain. Mr. Coursey was also an active experimenter in the early development of wireless telephony, and was the author of several books on this and allied subjects. He became chief engineer of the Dubilier Condenser Co., Ltd., in 1923, a position which he held until 1931, when he was appointed technical director of the Company; he retained that position with distinction until his retirement in 1957, when he was appointed technical consultant in addition to retaining his directorship.

Mr. Coursey was the originator of many important developments and held numerous patents. His extensive knowledge and experience have been a valuable contribution to the industries with which he was connected. He also took a prominent part in and made valuable contributions to the deliberations of the electrical and telecommunications industries both on British and international committees.

NEWS and VIEWS

Theoretical Physics at St. Andrews :
Prof. R. B. Dingle

Dr. Robert Balson Dingle, reader in theoretical physics in the University of Western Australia, has been appointed as the first occupant of the new chair of theoretical physics in St. Salvator's College of the University of St. Andrews. After a notable undergraduate career as Bournemouth Borough Scholar and major scholar at St. John's College, Cambridge, during which he held the Wright Prize twice and also the Hughes and Hockin Prizes, and at the end of which in 1946 he confused his Tripos examiners by, in effect, dismissing their questions and setting and answering his own, he found a corner in the Royal Society Mond Laboratory and began his research career, shortly thereafter being appointed to a fellowship at his College. Almost immediately, he began to publish, and a steady stream of valuable papers has continued since, uninterrupted by a series of moves of increasing geographical amplitude, first to Bristol, then Delft, then Ottawa, and then Nedlands in Western Australia.

Dr. Dingle's publications have covered three main fields and a few small ones. First, chronologically, came a series of elegant papers clarifying the hydrodynamics of liquid helium II, and in particular, second sound. This was followed by a series on collective electron effects in metals, in which diamagnetic or cyclotron resonance was predicted. This series also contained several important contributions to the understanding of the complicated oscillatory magnetic phenomena in metals known as the de Haas–van Alphen effect, and the removal of the apparent discrepancy between observation and theory of optical reflectivity in metals by an extension and correction of the Reuter–Sondheimer theory. The most recent series of papers is more mathematical and is concerned with the evaluation of integrals used in mathematical physics. In addition to these three main streams, he has also made useful contributions on semi-conductors and on quantum and statistical mechanics.

Pharmacology in London :
Prof. G. V. R. Born

Dr. G. V. R. Born, recently appointed to the Vandervell chair of pharmacology at the Institute of Basic Medical Sciences in the University of London, took his M.B. and Ch.B. at Edinburgh in 1943 and his D.Phil.(Oxon) in 1951. After his return from service as army pathologist in the Far East, he took up research in the rapidly developing new field of biochemical pharmacology. While on the staff of the Medical Research Council's Toxicology Research Unit he became interested in metabolic inhibitors, and, on returning to the Nuffield Institute for Medical Research at Oxford in 1953, he studied the actions of these inhibitors on smooth muscle. His main field of work has been on the pharmacologically active amines, and this led him to study the blood platelets by biochemical and pharmacological methods. A most notable contribution was the discovery of large amounts of adenosine triphosphate in platelets. The role of this substance in the uptake and storage of 5-hydroxytryptamine by platelets and its fate during clotting are now being explored. Dr. Born has also taken an active part in the teaching at Oxford, both in the Departments of Pharmacology and Pathology, and as a medical tutor at St. Peter's Hall. He has travelled widely and is well known to his scientific colleagues on both sides of the Atlantic.

College of Advanced Technology, Birmingham :
Dr. T. Lupton

Dr. T. Lupton has been appointed head of the Department of Industrial Administration of the College of Advanced Technology, Birmingham, as from April 1. At present senior lecturer in industrial sociology in the University of Manchester, Dr. Lupton began his career as an apprentice marine engineer, and later became an engineering draughtsman. He afterwards gained a diploma in economics and political science at Ruskin College, Oxford, proceeded to an honours degree in economics at Oriel College, Oxford, and was awarded the degree of Ph.D. at Manchester in 1959. Dr. Lupton has carried out a number of investigations on the relationship between occupation and social status, and recently the Department of Scientific and Industrial Research granted him £29,000 for a five-year research project on "Social Factors affecting Workshop Behaviour"; he will be continuing this work at Birmingham with the aid of Research Fellows and assistants.

National Institute of Agricultural Botany :
Mr. G. W. G. Briggs

Mr. G. W. G. Briggs has been appointed assistant director of the National Institute of Agricultural Botany. Since 1948 he has been head of the Seed Production Branch of the Institute, where he was responsible for the development of field approval and certification schemes for the improvement of cereal, herbage and vegetable seeds. The post of assistant director was created following the resignation of Mr. A. Sandison, who has been deputy director of the Institute during the past seven years.

Institution of Electrical Engineers: New Honorary Members

THE Council of the Institution of Electrical Engineers has elected the following to honorary membership: Colonel B. H. Leeson, recently director of the British Electrical and Allied Manufacturers' Association, in recognition of his extensive contributions to the application of electrical science and engineering in the manufacture of electrical plant and equipment, to the development of co-operative effort within the electrical industry and to the training of electrical engineers, and of the services he has rendered to the Institution during nearly half a century; and Dr. Richard L. Hearn, for his notable contributions in the development of electricity generation and its supply for the benefit and use of mankind, particularly in Canada, and for the part he has played in international co-operation in this field. Dr. Hearn was elected under the provisions of the rule whereby the Council is empowered to elect, once in every three years, an additional honorary member who is not at the time of his election a member of any class of the Institution.

Faraday Medal

THE thirty-eighth award of the Faraday Medal of the Institution has been made to Sir George Thomson, president of the British Association, in recognition of his outstanding contribution to the advancement of electrical science, of the leadership in research which he has given to others in this field, and of his inspiring services to education in science and technology. This Medal is awarded not more frequently than once a year, either for notable scientific or industrial achievement in electrical engineering or for conspicuous service rendered to the advancement of electrical science, without restriction as regards nationality, country of residence, or membership of the Institution.

Youth Services in Great Britain

IN the House of Commons on February 3, the Minister of Education, Sir David Eccles, announced that the Government accepted in principle all the main recommendations for Government action of the report of the Committee appointed in November 1958 under the chairmanship of Lady Albemarle to review the contribution which the youth service in England and Wales could make in assisting young people to play their part in the community. The necessary discussions between the Ministry of Education, the local education authorities and the voluntary youth organizations, with the view of taking precise decisions how to act on the Albemarle recommendations, will take place urgently. The Government has decided that deficiencies in voluntary and paid workers, in buildings and facilities for sport, in money and enthusiasm, must be made good, though everything cannot be done at once, and the will to succeed must come from the local areas at least as much as from the centre. The Government will finance by Exchequer grant most of the recommended expansion in training facilities from 700 to 1,300 full-time youth leaders by 1966, and Sir David hopes an emergency training college will be open by January 1, 1961. He proposes to ask interested parties to examine with him the execution of the recommendation regarding salaries and opportunities for transfer to other careers. Grants for starting buildings by the local authorities and voluntary organizations in the next two years will be authorized up to £2 million, and grants to voluntary projects will be increased. Grants will be increased for headquarters expenditure of voluntary organizations for approved experiments in attracting young people not now using the youth service. Sir David said that, to begin with, he proposes himself to take the chair of the recommended Youth Service Development Council.

Sir David undertook to consider a suggestion that the House should discuss the Albemarle and the Crowther Reports together, and said that the Government proposes to accept the recommendation that the age-range of the youth service should be 14–20 years. A corresponding statement was made on the same day by the Minister for Science, Lord Hailsham, in the House of Lords.

Gasification of Coal and Oil

REPLYING to a question in the House of Commons on February 1, the Minister of Power, Mr. R. Wood, said the Gas Council is spending £120,000 this year on research into coal gasification, and the National Coal Board and the Central Electricity Generating Board £680,000 and £130,000, respectively, on coal utilization. The Gas Council is spending about £230,000 on research into the gasification of oil under a programme intended to contribute also to the improvement of the economics of coal gasification, and another £240,000 on research into problems of gas purification and the treatment of by-products.

Trans-Pacific Telephone Calls

THE United Kingdom Government has accepted the report of the Pacific Cable Conference, which was held in Sydney during September–October 1959, and in which Government representatives from Canada, Australia, New Zealand and the United Kingdom participated. Cable and Wireless, Ltd., will act for the United Kingdom Government in the construction, maintenance and financing of the cable. The total estimated cost is £26·3 million, of which the United Kingdom share is £8·3 million. The cable will be more than 8,000 nautical miles long and will include more than three hundred under-sea amplifiers. In places, cable and amplifier will be laid to a depth of almost four miles. The project is to be undertaken by a joint Commonwealth Management Committee. The Committee will be under the leadership of Mr. R. J. Halsey, director of research of the United Kingdom Post Office. A light-weight cable will be used with rigid repeaters developed in the United Kingdom and being used in Cantat, the cable between the United Kingdom and Canada. The Trans-Pacific Cable will have a capacity for at least eighty simultaneous conversations. Any one of the telephone circuits can be used to provide twenty-four telegraph channels.

National Central Library

THE forty-third annual report of the Executive Committee of the National Central Library, covering the year ended February 28, 1959, records that although the requests for loans continue to become more specialized and inaccessible in nature, the proportion of applications handled successfully continues to rise and is now 76 per cent (National Central

Library. 43rd Annual Report of the Executive Committee for the year ending February 28, 1959. Pp. 20. London: National Central Library, 1959). Attention has been given to the problem of increasing the speed with which books can be obtained, and here delays are chiefly due to the failure of some libraries to deal promptly with the requests of the National Central Library. The Executive Committee appeals strongly to all co-operating libraries and regional bureaux to help by dealing with requests with the minimum of delay. Total applications again decreased, from 79,971 in 1957–58 to 77,004, and of the 76,048 volumes issued from or through the Library (including renewals), 32,666 were to or from public libraries; 12,202 to or from university libraries; special outlier libraries, 8,704; overseas libraries, 3,077; Government departments, 2,528; industrial and research organizations, 3,497; and adult classes, 11,286. Overseas libraries lent 1,804 volumes and borrowed 2,488 volumes, and 240 and 432 further requests, respectively, were satisfied by photographic reproductions. The outlier libraries, now numbering 281, lent 14,525 volumes, and other special libraries a further 2,527 volumes. Regional co-operation covered 163,842 loans within systems, and 22,099 outside systems and the university libraries supplied 24,390 books and borrowed 12,202. The arrears in the incorporation of entries for the union catalogues increased and the problem has been specially examined. There was a marked increase in the work of the British National Book Centre.

Teacher Training in Britain

More than 31,000 men and women are now training to become teachers, an increase of nearly a third compared with the 24,500 under training five years ago. The number entering training colleges this autumn had also increased by a third—16,000 compared with 12,000 in 1955–56. Standards are rising, most of the students now having more than the minimum academic qualifications for entry. The training colleges will thus be sending a record number of trained teachers into the schools in 1961. This will help to tide over the so-called 'year of intermission' in 1962, when there will be fewer newly trained teachers entering the schools as a result of the lengthening of the training course from two to three years. Expansion of the teacher training colleges by nearly two-thirds is now under way; many of the 130 training colleges are to be extended and new colleges built.

Recruitment for next year's intake is already starting, and the Ministry of Education has issued a revised booklet, "Becoming a Teacher", to help young people who want to teach in primary or secondary schools. The booklet describes the work the teachers do, the schools in which they teach, the qualifications they require and the training they undertake. It includes information about the training grants available and the salaries paid to teachers. The booklet can be obtained free from any local authority or from the Ministry of Education, Curzon Street, London, W.1.

Rubber Industry Display at the Science Museum

A permanent display representative of both producing and manufacturing sides of the British rubber industry has been presented to the Science Museum, London. It occupies three large show-cases in the industrial chemistry gallery, and illustrates the molecular structure of natural and synthetic rubbers; their production, compounding, and processing; the great variety of products thus obtained, and the vital part they play in modern life. The display has been devised and organized by the Federation of British Rubber and Allied Manufacturers and made possible by the generosity of British manufacturers of rubber products; makers of synthetic rubbers, compounding chemicals, processing machinery and components; the Natural Rubber Development Board, representing producers of natural rubber; and the National College of Rubber Technology.

National Museum, Bloemfontein

The annual report of the National Museum at Bloemfontein for the year ended March 1959 records the installation of a model of a gold mine and a dioramic display of a Bushman's cave showing rock paintings, stone implements and two modelled figures of men, one in the act of painting against the rock wall, while the other is making stone implements. In contrast with other museums, it was found that an admission charge curtailed severely the number of visitors, and after an experimental period of six months the charge was withdrawn. Much research work by the staff is in progress or has been completed. This includes studies on fossil hippopotami, engraved Bushmen sticks and Karoo vertebrate fossils.

Collecting Fossils, Minerals and Rocks

The seventh edition of "Instructions for Collectors", No. 11, published by the British Museum (Natural History) at 1*s*., follows the usual lines of this well-known and excellent booklet. It will be found to be of use to all types of collectors as, in addition to very elementary instructions, details are given of preparations which have only recently been discovered. The new materials mentioned have in all cases been tested extensively and may be used with confidence.

Marconi International Award

The establishment of an annual award in memory of Guglielmo Marconi is announced by Permindex, the World Trade Centre in Rome. This award will be made to the person making the greatest contribution to the development of Marconi's discoveries. Recipients will be chosen by a committee composed of important international personalities, a member of the Marconi family and representatives of Permindex. This announcement followed the dedication of the Marconi obelisk, to be known as the Marconi Antenna, in the Piazza Italia around which are grouped the four majestic buildings shortly to be occupied by Permindex. To celebrate the occasion the Square itself was renamed Piazza Guglielmo Marconi. The prize consists of a small reproduction in gold of the obelisk. Further information can be obtained from Industrial Trade Fairs, Ltd., Drury House, Russell Street, Drury Lane, London, W.C.2.

Ramsay Memorial Fellowships in Chemistry

The Trustees of the Ramsay Memorial Fellowships Trust will consider in June applications for two fellowships for advanced students of chemistry. One of the fellowships will be limited to candidates educated in Glasgow, who can apply to be considered for either fellowship. The value of each fellowship will be £600 per annum, to which may be added a grant for expenses of research not exceeding £100 per annum. The fellowships will normally be tenable for two years. Further information can be obtained from the Joint Honorary Secretaries, Ramsay Memorial Fellowships Trust, University College London, Gower Street, London, W.C.1. Completed application forms must be received not later than April 11, 1960.

Lady Tata Research Scholarships and Fellowships

The Trustees of the Lady Tata Memorial Trust invite applications for grants, scholarships and fellowships for research on diseases of the blood, with special reference to leukæmia, in the academic year beginning on October 1. In view of the affinity between leukæmia and other forms of neoplastic disease, candidates with programmes of research on any aspect of malignant disease which may throw light on problems of leukæmia will be eligible for consideration. Further information can be obtained from the Secretary of the (European) Scientific Advisory Committee, Lady Tata Memorial Trust, Chester Beatty Research Institute, Fulham Road, London, S.W.3. Applications must be submitted before March 31, 1960.

Lectures at the Imperial College, London

The nine inaugural lectures delivered at the Imperial College of Science and Technology during 1956–57 and 1957–58 have now been issued as a bound volume. They comprise those of Prof. B. G. Peters on "Parasitology at Imperial College"; Prof. J. G. Ball on "Metallurgy—Art, Technology, Science"; Prof. W. K. Hayman on "Pure Mathematics at a Technological University"; Prof. Abdus Salam, "Elementary Particles"; Prof. P. V. Danckwerts, "Chemical Engineering in a University"; Prof. D. H. R. Barton, "Some Modern Trends in Organic Chemistry"; Prof. C. C. Butler, "Ultra-High Energy Nuclear Physics"; Prof. F. D. Richardson, "The Changing World of the Extraction Metallurgist"; and Prof. J. M. Kay on "The Nuclear Power Programme and its Impact on Engineering Education" (pp. ii+134+24 plates. London: Imperial College of Science and Technology, 1959).

University News: Birmingham

The University of Birmingham and the University College of Rhodesia and Nyasaland have agreed that when a Medical School is established in the University College at Salisbury, Southern Rhodesia, this School will initially be an affiliated institution of the University of Birmingham. This will mean that, for the period of affiliation, students in the Medical School of the University College will be prepared for degrees in medicine of the University of Birmingham.

The following lecturers have been appointed: J. C. Maltbaek (chemistry); Dr. D. S. Urch (chemistry); G. C.-Y. Hu (highway and traffic engineering); Dr. C. G. Wall (chemical engineering); G. H. J. Bennett (industrial metallurgy); R. S. J. Good (mechanical engineering).

Cambridge

Dr. Kenneth McQuillen, University lecturer in biochemistry, has been appointed College lecturer in biochemistry in Churchill College as from October 1961.

Durham

The University of Durham has received a grant from the University Grants Committee to enable it to place orders for nearly £40,000 worth of additional equipment for the University Computing Laboratory at 1 Kensington Terrace, Newcastle upon Tyne 2. At present, the Laboratory is equipped with a Ferranti Pegasus electronic digital computer, and this installation is to be enlarged by the addition of magnetic tape equipment and of a fast output punch.

London

Dr. R. G. Lipsey has been appointed to the University readership in economics tenable at the London School of Economics and Political Science. The title of reader in economics in the University of London has been conferred on Dr. K. J. Lancaster in respect of his post at the London School of Economics and Political Science.

Announcements

The Messel Medal, the senior award of the Society of Chemical Industry, which is awarded every two years in gold, will be presented to Viscount Chandos in Bristol on July 6 during the annual meeting of the Society. The Medal is awarded for meritorious distinction in science, literature, industry or public affairs.

Prof. E. J. Wayne, regius professor of practice of medicine in the University of Glasgow, has been appointed chairman of the Clinical Research Board, in succession to Sir Geoffrey Jefferson.

Dr. David K. Detweiler, associate professor of veterinary pharmacology in the University of Pennsylvania, has been awarded a grant of 1,100,000 dollars over a period of ten years from the National Heart Institute of the National Institutes of Health, for research and training in comparative cardiovascular studies.

The International Society of Tropical Ecology, with its headquarters at Allahabad, India, was inaugurated by His Excellency the Governor of Bombay on January 5. The following officers were elected for 1960–61: *President*, Dr. J. C. Sen Gupta (Calcutta); *Vice-presidents*, Dr. F. R. Fosberg (Washington), Dr. C. G. G. J. Van Steenis (Holland); *Treasurer*, Dr. R. Misra; *General Secretary*, Dr. G. S. Puri.

An international symposium on "Inhaled Particles and Vapours", organized by the British Occupational Hygiene Society, will be held in Oxford during March 29–April 1. Further information can be obtained from Dr. J. S. McLintock, Medical Service, National Coal Board, Hobart House, London, S.W.1.

A symposium on "Inorganic Chemistry" will be held in the Wallberg Building, Department of Chemistry, University of Toronto, on April 8. Further information can be obtained from Dr. A. D. Allen, Department of Chemistry, University of Toronto, Toronto 5.

A symposium on "Continuous Culture of Microorganisms" will be held during March 31–April 1 at University College, London, under the auspices of the Microbiology Group of the Society of Chemical Industry. Further information can be obtained from the Assistant Secretary of the Society, at 14 Belgrave Square, London, S.W.1.

In the review of the book, "Mathematical Methods of Operations Research", by T. L. Saaty, published in *Nature* of December 5, 1959, p. 1763, it was stated that a printing error occurred on p. 340. Mr. R. Syski, of Hivac, Ltd., South Ruislip, Middlesex, has pointed out that this is not the case, and Prof. L. S. Goddard agrees, adding that the formula is repeated incorrectly on p. 346, where the exponent of μ/λ in the second term is given as $i + n - 1$, whereas the correct exponent is $i - n + 1$.

THE NATIONAL MUSEUM OF WALES

DEPARTMENT OF INDUSTRY

A DEPARTMENT of Industry was founded in the National Museum of Wales, Cardiff, in 1958, and its gallery was opened to the public last December.

The purpose of the Department is to illustrate not only the history and development of the older Welsh industries that were founded upon Welsh raw materials or upon techniques evolved or developed in Wales but also the application of science and research to the industries more recently introduced. In presenting an objective record of technical progress an attempt will also be made to show something of its impact upon human life.

The exhibits are displayed in fixed cases of shop-window type and in standard museum cases. For the present, there is a preponderance of those dealing with the iron and steel industry in Wales; in some exhibits it is looked at from the historical point of view alone, whereas in others the contrast between old and new methods is brought out by combining models of obsolete plant and photographs of modern processes.

The first of the iron-making exhibits contains a model of an anthracite blast-furnace used in the Gwendraeth Valley about 1850, and one of a blast furnace with a stove for heating the air before its injection into the furnace. The latter was associated with the successful attempts of John Budd in adding pre-heating chambers to some anthracite-burning furnaces at Ystalyfera in the Swansea Valley in 1844. Like other contemporary blast furnaces, those at Ystalyfera were constructed of massive masonry enclosing and supporting the fire-brick chambers in which the reactions took place. A further typical example of the construction adopted in South Wales before 1850 is depicted in a model of a blast furnace at the Pen-y-Darren Ironworks. Included in the model is a small pipe stove, consisting of nine semi-circular cast-iron pipes placed in parallel in a chamber heated by a coal fire, by which some of the blast was heated. Coupled with this model is one of a blowing engine used at the works for supplying air to blast furnaces. The type of blast furnace erected at Cardiff between 1888 and 1895 is represented by a half-section model. These furnaces were hand charged until 1927.

Obsolete steel-making plant is illustrated by a model of an open-hearth steel furnace of a type used in South Wales from 1890 until 1900, and included in the same exhibit is a sectional model of a Siemens' gas-producer, formerly the property of Sir C. W. Siemens, representing the modified type of gas-producer which he devised for use with the open hearth process perfected by him in 1867. In an exhibit on steel-making by the Bessemer process, there is a sectional model of a Bessemer converter of 1856.

In 1898 the works engineer at Dowlais Works designed and patented an ingot-stripping machine. Previously the cast-iron moulds had been stripped from steel ingots by hydraulic and steam cranes working alongside the sunken teeming pits. The new stripping machine, hydraulically driven, made it possible for ingots to be cast on bogies and then taken to the soaking pits some distance from the steel-making plant. A model of this machine is on view, and there is also a model of the original engine used at the same works for driving the roughing rolls of the rail-mill.

The hand processes of making tinplates in works dismantled during recent years are illustrated in an exhibit containing tools, photographs and samples of plates in the various stages of rolling from the original tin bar to the finished pack of eights.

At one end of the gallery there is a small transport section showing models of nineteenth-century sailing ships, illustrating the transition from sail to steam, and of early twentieth century cargo steamers under Welsh ownership. There is a model of the first Garratt articulated locomotive built for use in Great Britain, which operated at the Hafod Copper Works, Swansea, and one of a Taff Vale Railway engine and tender of 1867. A working model of a high-pressure steam engine designed by Richard Trevithick is also included; it is of a type installed at iron-works at Pen-y-Darren, Hirwaun and Tredegar at the beginning of the nineteenth century. A small panel of photographs alongside this model illustrates parts of the remains of the tram-road from Merthyr to Abercynon along which the Trevithick locomotive travelled in 1804.

One of the fixed cases contains a model of a rotary cement kiln. This will be supplemented by an exhibit adjacent to it which will include specimens of the raw materials used in the making of cement, of the product at various stages of manufacture, and of photographs of quarries and parts of the plant involved. An exhibit of Welsh exposed-aggregate slabs is also planned. A collection of tools, photographs and drawings illustrates the preparation of roofing slates in North Wales, and there is an exhibit devoted to the history and development of the miner's safety lamp.

The rayon industry is represented in an exhibit illustrating the manufacture of rayon staple and box spinning textile yarn at a factory in North Wales. An exhibit dealing with the nylon industry is being planned, and it is hoped to have another featuring the nickel industry.

Some original machines—a beam engine for pumping water from pits, a water-balance pit-head gear used to raise coal, and a set of rolls and a shear-table from an old-type tinplate works—are installed in the grounds of the Museum.

The gallery as it now stands indicates the field which the Department of Industry is intended to cover. It will be added to as rapidly as circumstances permit; but full use of models and original objects now in reserve cannot be made, nor can the full illustration of the special industries of Wales be achieved, within the confines of the present gallery. Much of this work will have to await the hoped-for addition to the Museum of a west wing.

WATER RELATIONSHIPS OF PLANTS IN ARID AND SEMI-ARID CONDITIONS

THE part played by Unesco in stimulating interest in the problems of the drier regions of the surface of the Earth is widely recognized. Its excellent series of symposia, covering hydrology, utilization of saline water, plant ecology, human and animal ecology, wind and solar energy, and micrometeorology, has done much to crystallize our current concepts of water as an ecological factor and particularly to emphasize the physical background in which it operates. These discussions were extended to the level of the individual plant at a symposium organized in Madrid during September 1959.

Considered as a whole, the two published volumes of the Canberra symposium on micrometeorology and the two volumes of the Madrid symposium, which have yet to appear, provide not only one of the best modern treatments of plant–water relations but also illustrate clearly the way in which plant ecology is casting its traditional shackles and emerging as an active experimental discipline. The Madrid symposium, although leavened with a few soil physicists and plant physiologists, was numerically dominated by plant ecologists. Among these were included representatives of the three modern categories: (i) the 'chroniclers', whose penchant for description and cartography readily translates suggestions into irrefutable dogma and leads to the creation of beautiful but often meaningless maps—multivariate analysis apparently does not yet qualify for an expense account; (ii) the 'partialists', whose methods have not advanced beyond the era of Livingston and Maximov, and whose thought is restricted to the confines of the plant; and (iii) the 'totalists', whose concern is with the entire soil–plant–atmosphere system. It is no accident that the modern approach to experimental ecology represented by the third category should have developed within the applied science of agriculture but simply reflects the stimulus arising from close association between people investigating detailed aspects of different parts of the same system. One of the finest examples of an ecological investigation is that reported at Madrid by R. O. Slatyer on the water relations of two natural communities in Australia. Here, soil physicists, plant physiologists and micrometeorologists are working together and each has a full appreciation of the complete system.

It is also no accident that this approach should emerge first with the study of water because the responses of the plant to water are less complex than to other ecological factors such as light or temperature. Knowledge of the control of the water supply has now advanced to the stage of complete quantitative description, although the values of some parameters are far from exact. Those which are least perfectly known include moisture-release curves, surface roughness, stomatal conductance and the variations in extent of the root-zone with time, species and environment. Several contributors discussed these. The location and sizes of the resistances in the plant to liquid flow are uncertain, although it is now clear that plants may experience an adverse deficit of water in the leaves with the soil water at 'field capacity' when the potential transpiration exceeds ~ 4–5 mm./day. In order to evaluate this and other problems intelligently and to remove general confusion, the status of water in all parts of the soil–plant–atmosphere system must be expressed in equivalent terms. The logical terminology is that of thermodynamics, and all members of the conference unanimously agreed to use energy terms and units henceforth. The *water potential* (difference in free energy between the water contained in the soil or plant system and pure free water at atmospheric pressure) should be used for the quantity normally associated with total soil-moisture stress and diffusion pressure deficit or suction pressure. It is determined by three components: (1) the *osmotic potential*, which takes account of all dissolved materials in the soil solution or plant sap and is usually called the osmotic pressure; (2) the *pressure potential*, which is associated with the total pressure difference in the system, that is to say, with the difference in pressure between the water in a tensiometer and that in the adjacent soil or with the turgor pressure of plant cells; and (3) the *matric potential*, which depends on the solid matrix and the forces of retention of water in the matrix and which has been associated with terms previously called soil suction or moisture tension but which is usually ignored in plant systems.

Knowledge of the response of plants to water supply is in a less happy position than is our appreciation of transpiration. The differing responses between species, and within the one species with phase of growth, to the apparent soil–water deficit arise mainly from differences in the rate of extension of the root system, from measuring the deficit at one or two depths only, and from the difficulties associated with obtaining an integrated value of the water deficit in the soil actually occupied by roots. With increasing values of the water deficit in soil fully permeated by roots at any point of time, growth appears to decrease over the range of water potential from $\sim -0{\cdot}01$ ('field capacity') to $\sim -1{\cdot}5$ joules/gm. ('wilting point'), but the form of the relationship is modified by the level of potential transpiration and other environmental aspects. R. M. Hagan, N. S. Petinov and S. A. Taylor discussed irrigation within this context.

Survival under dry conditions—the problem of drought resistance—is still little understood. The mechanisms involved, discussed by H. R. Oppenheimer, A. M. Migahid and other contributors, are complex and numerous. Some of these, such as extensive root systems, leaf fall and stomatal closure associated with low cuticular conductance, prolong the period during which the water supply of tissue capable of growth can be maintained. The reaction of cells and tissues to drying, analysed by O. Stocker, also differ, and the behaviour of the whole plant is related to the resultant of the complex interactions which occur between varying proportions of cells of differing responses. One of the most exciting developments in this field is the hardening of plants prior to sowing which has been developed by Soviet scientists.

Increases in yield from 10 to 40 per cent are claimed. These and other contributions from the U.S.S.R. to the understanding of the reactions of plants to water and to heat were discussed by N. S. Petinov and P. A. Genkel.

The discussions will be published in two parts: (1) Reviews of research prepared as background material on "The Income and Loss of Water" (F. L. Milthorpe), "Soil Water Relations" (W. R. Gardner), "Physiological and Morphological Changes in Plants due to Water Deficiency" (O. Stocker), "Adaptation to Drought—Xerophytism" (H. R. Oppenheimer), "Methods of Research on Water Relations" (F. Eckardt), "The Management of Native Vegetation" (R. M. Moore), "Principles of Dry Land Crop Management" (S. A. Taylor), "Significance of Fallow as a Management Technique" (W. J. Staple), "Principles of Irrigated Cropping" (R. M. Hagan and Y. Vaadia), "Survey of Research on the Water Regimes of Plants" (P. A. Genkel and K. A. Bodanova), "The Water Regime of Plants on Saline Soils" (B. P. Strogonov); (2) Proceedings of the symposium, which include about forty papers contributed on detailed aspects within the above fields by workers from seventeen different countries.

F. L. MILTHORPE

MINING GEOPHYSICS

THE second Nordic Meeting for Mining Geophysics was held at Haandværkerforeningen, Copenhagen, during December 7–8. More than fifty geophysicists and technicians, including five from Finland, eleven from Norway, and eighteen from Sweden, attended. The sessions were presided over by Haakon Brækken from Trondheim, Norway, chairman of the planning committee.

The technical programme included twelve papers, two of which were concerned with Greenland. B. Sjogren (Sweden) reported on a seismic refraction and reflexion investigation on the Malmberget glaciers. The area surveyed lies about 20 km. south-east of Mestersvig, Kong Oscars Fjord, east Greenland, and the purpose of the survey was to determine the thickness of the ice–moraine layer of the glaciers. The bed-rock is mainly sandstone from the Carboniferous period which is superseded by tertiary granite. The bed-rock velocities measured varied from 4,800 to 5,400 m. per sec. while the longitudinal wave velocity in the ice–moraine layer was between 3,700 and 3,800 m./sec.; the transverse wave velocity in this layer was of the order of 1,600–1,700 m./sec. The measured thickness of the ice–moraine cover varied from 120 to 450 m. The bed-rock under the glaciers was discovered to be U-dale shaped with relatively shallow slopes while the rock walls on the sides of the glaciers climbed very steeply to about 700 m. above the glacier surface. The second paper dealing with Greenland, "A Study of the Disturbances due to Salt-water on Minor Loop-frame Indications", was presented by J. Bondam (Denmark). The survey is part of experimental measurements carried out near Narssaq, south-west Greenland.

The important problem of the elucidation of structure was illustrated by G. F. Sakshaug (Norway). Electromagnetic, magnetic, and self-potential surveys have been employed in the exploration of copper mineralization at Bidjovagge, Finmark (Northern Norway). Electromagnetic surveys have been conducted as a general reconnaissance method as well as towards the examination of detail. Electromagnetic anomalies are largely confined to beds of graphite shale occurring in the area. Magnetic surveys define magnetite-bearing greenstones. Self-potential surveys give essentially a map of the graphite shale under the moraine cover. All three methods have been essential to the interpretation of the structural relations in the area. Irregularities in the anomalies reflect tectonic disturbances, and ore-mineralizations—as diamond drilling has confirmed—are related to areas of intense disturbance. Another example of structure mapping was given by V. Munther (Denmark) in a presentation of a magnetic survey of Bornholm and its application to the geology, especially the tectonics. The island has been covered by magnetic stations in squares, the side-length being 500 metres. Moreover, a considerable number of traverses has been established, stations distances being from 10 to 20 m. apart. It has in this way been possible to correct the previous boundary lines between granite and sandstone. Known faults have been traced in more detail and older faults have been shown. In the granitic area the magnetic contours indicate the granite variety. Interpretation of the anomaly map indicated partly stronger magnetic layers at a greater depth, partly a change in the magnetic field with time. The practical aspect of the survey is the economic interest in sandstone.

A case-history was demonstrated by J. Espersen (Sweden). Magnetic and gravimetric measurements have pointed out the existence of two important and several minor ore bodies at Svappavaary near Kiruna, north Sweden. The applicability of magnetometers and gravimeters in ore prospecting is underlined by the confirming drilling results. J. Nuutilainen (Finland) described gravimetric application in ore prospecting. Levelling is carried out by means of a mercury pressure-gauge and a water tube which at the same time is the measuring tape. H. Paarma (Finland) considered magnetic gradient measurements with regard to ore bodies and structures. A report of interest concerning the conductivity of magnetite, sulphide, and different rocks sampled in Swedish mines was given by Sture Werner (Sweden). Measurements were undertaken in fifteen sulphide-mines and twenty iron-mines. The resistivity, which is the reciprocal of the conductivity, was found to amount to 300,000–3,000,000 ohm-cm. for the circumjacent rocks, while the value for magnetite and sulphide was usually found to be of the order 0·01 ohm-cm. The resistivity increases rapidly as the percentage of magnetite decreases; for example, an ore sample with 50 per cent iron has a resistivity of about 10,000 ohm-cm. For the sulphide samples the resistivity increases slowly as the percentage of sulphide decreases. Only at a very low sulphide content do the resistivity values rise.

A new instrument for measuring the curvature in bore-holes was presented by A. Holm (Sweden), and A. Breen (Norway) discussed the employment of phase-sensitive voltmeters in measuring electromagnetic fields. A point of practical importance was reported by B. Karlemo (Sweden) in a description of methods for production of geophysical maps.

At a joint evening meeting with the Danish Geophysical Society, D. Malmqvist (Sweden) gave a review of electrochemical ore-prospecting methods and emphasized the utility of this prospecting method. The 'polarization provoquée' method proposed by Schlumberger about forty years ago seems to be very suitable, especially in cases dealing with disseminated ores, and Mr. Malmqvist demonstrated some survey examples from northern Sweden.

The chairman, H. Brækken (Norway), introduced at the business meeting the very important problem of education of mining geophysicists. The meeting appointed a committee (Brækken, Werner and Tengstrøm (Sweden)) to consider the different points and to report at the next meeting.

With regard to the planning committee for the third meeting, which is contemplated within two years, the meeting appointed B. Tornqvist as chairman (Sweden), and H. Brækken, M. Puranen (Finland), and S. Saxov (Denmark) as members.

In all, the second Nordic Meeting for Mining Geophysics proved to be most successful, the attendance being about 100 per cent higher than at the first meeting at Boliden, Sweden, during November 7–9, 1956; the discussions were many and the opinion of the meeting strongly supported the arrangements for future meetings and co-operation in common problems.

SVEND SAXOV

THE MEDICAL RESEARCH COUNCIL

REPORT FOR THE YEAR 1957–58

THE recent report of the Medical Research Council for the year 1957–58, prepared by the Committee of Privy Council for Medical Research, adopts once more the form now used for several years (Cmnd. 792. Pp. viii+288. (London: H.M. Stationery Office, 1959.) 13*s*. net). Instead, that is to say, of giving detailed accounts of all the researches in progress, it gives articles based on certain aspects of the Council's very varied programme, the intention being to deal, once every few years, with all the important fields of research undertaken by the Council's staff. These articles are also published separately under the title "Current Medical Research" (Pp. iii+46. (London: H.M. Stationery Office, 1959.) 3*s*. 6*d*. net).

In this publication and also in the report itself we are given a description of the Council's Laboratory in Gambia, West Africa. As part of its contribution to research in tropical medicine, the Council maintains three laboratories, in Uganda, Jamaica and Gambia, respectively, and that in Gambia here described is the oldest and largest. Unlike the others, it is self-contained and is not attached to any local institution. Situated at Fajara, it was originally part of the Council's Human Nutrition Research Unit in London, but its scope has now been widened to include research, not necessarily on tropical medicine, but in other fields as well. Many research workers use its increased facilities—cardiologists, chest and other specialists, pædiatricians, public health workers and others. The centre is visited by many research workers from Britain and has lately been used by a team representing the Council's Research Group on trachoma, a cause of blindness which, because of the menace it is, is engaging the attention of research workers all over the world. This centre is also developing research on the hyperendemic malaria of the Gambia region, a region which offers special facilities for work of this kind, and this is discussed in a separate article in the report. Much of it is done at Keneba, and to facilitate communication between Keneba and the laboratories at Fajara a radio-telephone has been set up, and the Wellcome Trust has generously provided a cabin-cruiser, *The Lady Dale*, which can also be used as a floating laboratory.

As well as the article just mentioned on the hyperendemic malaria of the Gambia area, the report includes articles on the remarkable work being done by members of the Council's staff on intracellular organelles and on the enzymes contained in them. Other articles discuss the work being carried out on the viruses of trachoma and inclusion blenorrhœa, the types of the bacillus that causes Sonne dysentery, the cancer-producing viruses and their immunology, the ultra-microscopic analysis of body fluids, the performance of coal miners in hot atmospheres, the fertilization of the mammalian egg and the rehabilitation of schizophrenic people. Another article of interest discusses research in general medical practice, a line of work which is much to be encouraged. An innovation in the report is the introduction of two photographs, one of which shows *The Lady Dale*, and two line illustrations.

For the rest the report contains its usual features. Brief summaries of the work of each of the Council's research units are given, together with the names and research addresses of the Council's many workers. A long bibliography lists the varied work that these workers have published

G. LAPAGE

COMPARISON OF RAIN-GAUGES

THE measurement of precipitation is a difficult problem because of the great variations in the nature of the precipitation and of the effects of wind eddies around the gauge. The radii of liquid drops and their rates of fall range from 0·2 mm. at 0·7 m./sec. of drizzle to 5 mm. at 8 m./sec. of the heaviest thunderstorm rain, while the speed of the wind around the gauge can vary over a somewhat greater range. Solid precipitation is even more difficult to measure. It ranges from fine snow to heavy fast-falling hail-stones and may at low temperatures completely choke the gauge if, as is usual, no heating is provided for melting. In addition to these inherent difficulties, rain-gauges must be cheap and simple to operate because, on account of the great variations in rainfall from place to place, it is necessary to have many of them widely distributed. Different types of gauge have been evolved in different countries in the endeavour to meet these difficult requirements.

The ordinary British type is fixed in the ground and has a catching cylinder with a diameter of 5 in., and which is set with the rim 1 ft. from the ground. Some Continental countries fix rain-gauges to posts so that the rim is 1 m. to 1·5 m. above the ground. The shapes of the interior of the cylinders differ considerably, while in some countries the catching cylinder is contained in a much wider coaxial cone called a shield intended to minimize the effects of wind eddies.

One of the basic principles of meteorological measurement is that observations, though their absolute accuracy is to some small extent inevitably uncertain, shall be comparable. With this aim in mind, the Royal Meteorological Institute of Belgium has recently completed a five years comparison of Belgian, British, German and French rain-gauges at the Uccle Observatory near Brussels and at Yangambi in the Congo. The work is described by Dr. L. Poncelet in a recent publication by the Institut Royal Météorologique de Belgique (Publications Série A. No. 10 : Sur le Comportement des Pluviomètres. Pp. 58. Uccle-Bruxelles : Institut Royal Météorologique de Belgique, 1959). The results are analysed according to season, intensity and continuity of fall, occurrence of sunshine after rain (because of evaporation of some of the catch), and wind speed and direction. The amounts collected in individual months vary greatly from gauge to gauge and a long period of averaging is needed to produce a consistent pattern of differences. Over the whole period the differences were less than 1 per cent, with the German and French high-level gauges giving lower readings than the low-level Belgian and British gauges. Dr. Poncelet considers that different types of gauge can be compared to an accuracy of 1 per cent, and that the relation of the annual totals to the precipitation which would actually reach the ground where the gauge stands during a year is of the same order of accuracy. The accuracy is appreciably less for snow at 2–3 per cent.

Dr. Poncelet, in his capacity as chairman of the World Meteorological Organization Working Group on the International Comparison of Precipitation Gauges, has also written an article in the October 1959 edition of the World Meteorological Organization's *Bulletin* on the use of a specially designed international reference precipitation gauge. This gauge is fixed in the ground and has its rim at a height of 1 m. and is contained in a conical coaxial shield the width of which at rim-level is 1 m. Arrangements are in hand for comparisons to be made in most countries between the readings of this reference gauge and the national gauges. The latter may, it is believed, differ from one to another by 5–15 per cent. Most of the reference gauges will be made by the same (British) manufacturer. In any event, every effort is being made to ensure uniformity down to the degree of polish of the copper surface and the wetting properties of the polythene receiving bottle and measuring glass.

It is hoped that this comparison, which will take several years, will provide means of reducing rainfall observations for comparison with an accuracy of 1–2 per cent, which will permit computations of water balance to be made and rainfall maps drawn without discontinuities at national frontiers.

G. A. Bull

MEASUREMENT OF VISUAL RANGE

By Dr. P. CROSBY

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TWO photoelectric instruments have been constructed at the Weapons Research Establishment, Salisbury, South Australia, for measuring the scattering coefficient, b, of the atmosphere. Because the associated 'visual range' V is more commonly understood, it is our practice to quote the results as 'visual range' values, where $V = 3{\cdot}912/b$ (see ref. 1).

Polar nephelometer. The first instrument is a version of Waldram's polar nephelometer[2]. The light source is an ionic flash tube. The light beam is defined by a baffle-box with square apertures. A second baffle-box with a photomultiplier tube forms the photometer. The apertures are square and the field of view of the photometer passes through the light beam. The photometer is rotated about a fixed point of the light beam to receive light scattered at various angles, from 15° to 165°. The volume of illuminated atmosphere that lies in the field of view of the photometer being known from the dimensions of the apparatus, a polar diagram of scattering of the light flux scattered by unit volume of atmosphere is obtained and extrapolated over the regions 0°–15° and 165°–180°.

The polar diagrams show a strongly forward scattering, indicating that most of the scattering is from large particles.

A measurement of the intensity of the source is made by directing the photometer at the source with a very dense neutral filter in the light-path. Integration of the polar curve produces a value of b or V.

The filter is required to prevent overloading the photomultiplier. Its transmission factor is calibrated against the reflexion of light from two magnesium oxide surfaces.

Integrating nephelometer. The second instrument is a version of the Beuttell and Brewer integrating nephelometer[3]. The light-source behind a piece of opal glass is an ionic flash tube. The photometer consists of a photomultiplier tube and baffle box with circular apertures. It is directed across the opal glass towards a black box in order to receive light scattered at angles between 10° and 170°. A correction of 6–7 per cent is required to allow for light scattered between 0°–10° and 170°–180°, according to the polar nephelometer results. The amount of light flux reaching the photomultiplier is directly proportional to the scattering coefficient.

A reference measurement of the intensity of the source measured along the normal to the opal glass is required to obtain absolute values of *b*. The intensity is measured by placing a diffuse reflector above the opal glass, on the axis of the photometer, to direct light through a dense filter on to the photomultiplier.

The photomultiplier output of either instrument is measured as the voltage across a 10^5-ohm anode load using a cathode-ray oscillograph with a long-persistence tube.

Meteorologists should be particularly interested in the integrating nephelometer since it is a direct-reading instrument. First, the voltage applied to the photomultiplier is adjusted to produce a standard cathode-ray oscillograph deflexion when measuring the intensity of the light source. Then, when the photometer is receiving scattered light, the 'visual range' is read directly by noting the cathode-ray oscillograph deflexion on the special scale placed over the screen. The instrument is 7 ft. × 18 in. × 18 in. and costs less than £200, excluding the cost of the two power supplies and the cathode-ray oscillograph.

Observations are made inside a well-ventilated room. Complete darkness is not required. Experience has proved that the results obtained indoors are identical with those outdoors within the 10 per cent accuracy claimed for the system, on dull days and at night-time. Checks during sunlight are not possible, as there is too much background stray light.

The visual range fluctuates widely during any one day. Generally, visibility is between 25 and 45 miles. Lower readings are obtained on occasions. The least was 0 5 miles during a mild sandstorm. The highest was 53 miles after heavy rain had fallen. The equipment is to be transferred to Woomera to study conditions there.

The 'visual range' is, according to meteorological definition, the distance at which a dark object can be distinguished and recognized against the horizon sky. It should be noted that seldom has a meteorological office any special markers and that generally a glance outside is the only basis of the 'visibility' figure published by the office. Sometimes an inspection of various distant landmarks is made, but estimates by different observers vary widely due in part to the different luminal contrast of their eyes. In any event, the ability to see a distant object depends on the prevailing background brightness in that direction, and the presence of cloud would affect the result.

The nephelometers, on the other hand, make a direct measurement of the basic property of the atmospheric scattering which is the primary cause of loss of contrast or brightness of an object as it recedes from the observer.

Special features of the new nephelometers compared with the visual instruments are:

(*a*) Use of an ionic flash tube. This produces a high-intensity light beam which need not be concentrated further by mirror or lens systems. The latter would have surfaces on which dust could collect and form stray light. The heat energy within the light beam from an ionic flash tube is not sufficient to dry the sample of atmosphere. Continuous sources, used in visual nephelometers, produce a great deal of heat.

(*b*) Use of photo-multiplier tube. This eliminates the fatigue of visual measurement of the low intensities of scattered light.

(*c*) Automatic recording of visibility. The visibility is read directly off the cathode-ray oscillograph screen of the integrating nephelometer after the calibration has been carried out. The light source can be operated automatically and a continuous photographic record of the screen made. Alternatively, the cathode-ray oscillograph can be replaced by a pen-recording system. In either case, a chart similar to a temperature-time chart is produced and would be valuable for forecasting visibility.

This communication is submitted by permission of the Chief Scientist, Australian Defence Scientific Service, Department of Supply, Melbourne.

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[1] Middleton, W. E. K., "Vision through the Atmosphere", 105 (Toronto Press, 1952).

[2] Waldram, J. M., G.E. Co. Ltd. Rpt. 8064, and Middleton (above), p. 201.

[3] Beuttell and Brewer, *J. Sci. Instr.*, 26, 357 (1949). *CDRC(G)* Report 52, and Middleton (above), p. 204.

IONIC EXCHANGE AND THE STRUCTURE AND FUNCTION, INCLUDING MOTILITY, OF CYTOPLASMIC MEMBRANES

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CONSIDERABLE study has thus far yielded no adequate unified explanation of the structure and function of superficial cytoplasmic membranes. Various metabolic, or carrier, hypotheses have not been substantiated when tested. Other chemical explanations, such as the lipoid – sieve hypothesis which views membranes as static structures, are limited to certain aspects of permeability. Our present increased knowledge of ionic exchange and the realization that macromolecular constituents of the cytoplasm are capable of functioning as ionic exchangers present the possibility of an explanation of membrane structure and function based on certain principles of ionic exchange.

A key problem is the formulation of an explanation of the conversion of the chemical energy of metabolism to cellular motions; one possible explanation is a reversible hydration – dehydration of the membrane. This sort of change can be an effect of certain types of ionic exchanges and may be influenced by a variety of compounds. As outlined below, this hypothesis, that ion-exchange phenomena may produce reversible hydration, permits a unified explanation of the electrical behaviour of cells, of differential permeability and of membrane motility.

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Because of the relatively recent applications of ionic exchange principles to biology, a few general points about them should be reviewed. There are two types of cationic exchangers: strongly and weakly acidic; and two types of anionic ones: strongly and weakly basic. Particles of ionic exchange materials consist of insoluble frameworks, permeable to water and with firmly bound acid or basic functional groups. Ion exchange depends on charge density: trivalent ions displace divalent ones, and so on. Concentration of ions is also a factor. Cross-linkage is the term for the rigidity of bonding within exchange particles. The specificity of exchange depends on cross-linkage and properties of the compounds making up the exchanging particles Rates of exchange decrease with increases in cross-linkages. Exchange-rates can be decreased by the binding of large ions on particle surfaces[1].

Biological materials may act as ionic exchangers[2]. Proteins, acid mucopolysaccharides and cerebrosides are weakly ionized, whereas the sulphated mucopolysaccharides, nucleic acids and phospholipids represent strongly ionized exchangers. Proteins exchange or bind ions such as hydrogen, calcium and the polyvalently ionized macromolecules. Fibrous proteins, long believed to form part of membrane structure, may also exchange hydrogen ions for sodium and potassium. However, the exchangers appear to work better in an insoluble form. Hence, it may be of interest that these biological exchange materials can exist in the cytoplasm as particles.

There seems to be good circumstantial evidence that the membranes are composed of some sort of cytoplasmic particle. It has been possible to demonstrate both the motility of cytoplasmic membranes and pinocytosis[3]. Certain cells have been shown to take up foreign proteins by phagocytosis, pinocytosis or both[4]. These processes are something of a puzzle, implying as they do not only a transient rupture of the cytoplasmic membrane but also a rather sudden increase in the amount of membrane material as well. If membrane material is potentially particulate and if the particles are drawn from the cytoplasm as needed, these puzzles can be solved. This implies that there are cytoplasmic particles that have some properties in common with membranes. Outstanding properties of membranes are permeability to 'fat-soluble' materials and electrical phenomena; the former implies a high content of lipid, whereas the latter implies ionic exchange because of an absence of metallic conductors. Microsomes have a high content of lipid and of nucleoprotein, so they may take a role in membrane formation. However, electron-micrographs which show the superficial cytoplasmic membranes and the mitochondria do not reveal any structures that can be clearly identified as microsomes[5,6]. Perhaps microsomes are in some measure created by the stress of high-speed centrifugation, and, if so, these particles and membranes may have a common precursor. Because membranes act as a lipoid phase, it is plausible that these particles form the membrane by coalescence of their lipids.

Assuming the existence of these membrane particles, one may indicate the effect hydration would have on permeability to water-soluble substances. Hydration of the lipids of these particles, if it went far enough, would increase permeability. This permeability would not only involve materials such as the sugars, but also the water-soluble ions. Under suitable conditions a flow of ions would produce the electrical phenomena. Hence, if one understood how to hydrate or dehydrate the membranes, one could exert some measure of control over both permeability and bioelectrical phenomena. Parenthetically, one should note that these particles would be in constant motion relative to the fluids which bathe them and that this could produce a situation like that in an ion-exchange column.

Hydration and dehydration of the cytoplasmic membranes (either by direct introduction or withdrawal of water or else simply by altering its activity) imply changing composition. Introduction of fat-soluble organic compounds could affect the hydration of membrane lipids. Carboxylic acids would tend to bind hydrogen and calcium ions and thus cause some dehydration. Phospholipids, being relatively highly ionized, would tend to be hydrating agents, and they could also tend to draw other hydrated ions into the membrane. Carbonyl and hydroxyl groups would tend to be hydrating agents whether they occurred in glycolipids or steroids. Organic ions, being fat-soluble and attended by water of hydration, would tend to be hydrating agents. In the opposite direction the more nearly non-polar ethers, esters, hydrocarbons and noble gases would tend to promote dehydration.

Reversible hydration is central to the explanation of membrane motility. Under suitable conditions the membrane particles would function as weakly acid cation exchangers. Such exchangers may function as converters of the chemical energy of ion-exchange to mechanical work because they undergo large increases in volume when they exchange their hydrogen for some other ion, and large decreases in it following a second treatment with acid[1]. The basis for such effects would be the carboxylic acid groups of the membrane particles. An increase of permeability would, for obvious reasons, tend to increase metabolism. Since acid, for example, carbon dioxide, is an important end-product of metabolism, the hydrogen ions formed would tend to produce a decrease of permeability and particle shrinkage. This change would be followed by a decrease in metabolism or less acid. The hydrogen ions in membrane lipids could then be exchanged for other cations in the environment, and this would lead to swelling of particles, increased permeability and so on. These changes would give rise to bioelectric phenomena.

A hydrated membrane, because of permeability to ions, would have a low electrical resistance and might be thought of as being depolarized. As acid from metabolism displaced cations from it, the membrane would become less hydrated and offer greater electrical resistance. The binding of hydrogen ions by the membrane would leave an excess of anions in the cytoplasm which would thus be negatively charged. Depolarization might be achieved by introduction of fat-soluble ions such as acetyl choline[7] into the membrane. Depolarization-levels of such ions would depend on the relative proportions of the various 'hydrating' or other agents in the particles.

The explanation of differential permeability is ordinarily to be subdivided into two parts: selective accumulation (or the reverse) of ions and of un-ionized substances. Differential permeability to ions may possibly be explained as an exchange or displacement process. The sort of mechanism involved (bearing concentrations in mind) may be illustrated by a displacement series of cations, which is arranged in order of decreasing ability to displace crystal violet from a weakly acid silicate exchanger, as follows: putrescine, cadaverine, pyridoxine, thiamine, histamine, hydrogen, barium, acetyl choline, calcium, magnesium, prostigmine, atropine, epinephrine, and esserine[7]. The arrangement follows the principles of ionic exchange and 'ion exclusion'.

The explanation of differential permeability to un-ionized materials requires the introduction of another idea from ionic exchange. This concept (absorption or ion-exclusion) is the absorption of slightly ionized or uncharged molecules by ion-exchanger particles. Some simple rules apply to this absorption. Aromatic acids are more strongly absorbed than aliphatic ones. Non-polar substituent groups enhance and polar ones diminish this absorption. Absorption processes have made it possible to separate isomeric six-carbon sugars from each other (borate exchangers) and, by means of exchangers with optically active functional groups, it has been possible to separate optical isomers[8]. Absorption, since it can help to account for the differential binding and exchange of optical isomers, aromatic compounds, polyhydroxy compounds and lipids *in vitro*, may account for the binding of lipids to a variety of multivalent macromolecular ions in the formation of cytoplasmic membranes. If, for example, it is possible to displace one hexose by another on a borate exchange column, it would be possible to displace one hexose by another in a membrane particle where the role of borate was taken by some other residue. This is a possible mechanism of differential permeability of cells to un-ionized water-soluble compounds and an alternative to the 'carrier molecule' hypothesis.

Proposing an alternative to the metabolically derived carrier hypothesis means considering metabolism to be only indirectly connected with membrane function. The action of cyanide on mammalian erythrocytes illustrates, experimentally, the limitation on the metabolic interpretation of cell function. The action of cyanide has been assumed[9] to be due to the inhibition, demonstrated *in vitro*[10], of cytochrome oxidase. However, cyanide has also been implicated as having an effect on cytoplasmic membranes; Brinley's *Amoebae* survived injection of a dose of cyanide that was lethal, extracellularly[11].

The effect of cyanide on active transport of cations and glucose utilization in erythrocytes during an 18-hr. incubation period was studied by Maziels[12]. While cyanide in concentrations up to 10 mM/l. did not inhibit active transport, the potassium – sodium ratio was smaller and the amount of inorganic phosphate liberated was greater in cells treated with cyanide than in controls at comparable pH levels. Also, the amount of glucose utilized was greater in the cells treated. The implication of an increase in general permeability was noted in the results of this work on glucose utilization, phosphate liberation and cation ratios. Since cytochrome oxidase is of no consequence in red blood cells, the probable effect of cyanide would be to bind hydrogen ions. In terms of my hypothesis, this would lead to increased permeability.

The indication for further study along this line is clear. As the ion-exchange hypothesis provides a better explanation than does the cytochrome-oxidase-inhibition theory for the action of cyanide on mammalian erythrocytes, so through 'ion exclusion' or absorption a better understanding of structure – action relationships in drugs may be possible. Indeed, in the general application of ion-exchange phenomena to aid our understanding of biology, we may be seeing the start of a rational approach to such fields as immunology, oncology and neuropharmacology.

[1] Pepper, K. W., and Hale, D. K., "Ion Exchange", 13 (Soc. Chem. Industry, London, 1955).

[2] Calmon, C., and Kressman, T. R. E., "Ionic Exchange in Organic and Bio-Chemistry" (Intersci. Pub., New York, 1957).

[3] Pomerat, C. M., *Ann. New York Acad. Sci.*, **58**, 1311 (1954).

[4] Schechtman, A. M., *Int. Rev. Cytol.*, **5**, 303 (1956).

[5] Sjostrand, F. S., and Hanzon, V., *Exp. Cell. Res.*, **7**, 393 (1954).

[6] Palay, S. L., and Palade, G. E., *J. Biophys., Biochem., Cytol.*, **1**, 69 (1955).

[7] Dettbarn, W. D., Wilson, I. B., and Nachmannsohn, D., *Science*, **128**, 1275 (1958).

[8] Wheaton, R. M., and Bauman, W. C., *Ann. New York Acad. Sci.*, **57**, 1950 (1953).

[9] Goodman, L. S., and Gilman, A., "Pharmacological Basis of Therapeutics", second edit., 931 (Macmillan, New York, 1955).

[10] Stotz, E., *et al.*, *J. Biol. Chem.*, **124**, 745 (1938).

[11] Brinley, F. J., *Proc. Soc. Exp. Biol. Med.*, **25**, 305 (1928).

[12] Maziels, M., *J. Physiol.*, **112**, 59 (1951).

SCRAPIE: A TRANSMISSIBLE HEREDITARY DISEASE OF SHEEP

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SCRAPIE is a chronic wasting syndrome affecting sheep in middle age, well known in Western Europe for at least two hundred years, almost always fatal and associated with degenerative changes in several organ systems. Its cause remains unknown; it can be transmitted experimentally by inoculation and some have considered the evidence available to favour a virus infection with a long incubation period[1], others a hereditary disease[2]. The literature has been reviewed recently[3].

The epidemiology• of scrapie has been studied continuously over a 7-yr. period in 70 flocks in Britain of 10 breeds with some 10,000 breeding ewes; many of the flocks are fully recorded and are closed populations except for new sires[2]. Some clinical and pathological criteria of the disease have been described[2]. Only unequivocal cases of classical-type scrapie are included in this study, which is based on 800 cases, of which the great majority were in females, only 40 being in rams. 149 cases have been autopsied. Affected animals have not been isolated in quarantine, but their progeny and close relatives have been disposed of so far as has been practicable.

The results may be summarized as follows:

(1) *Age of Manifestation.* Data are available for 526 animals—492 females and 34 males. The age of manifestation varied from 1¾ to 11 years in females and 1¼–5½ years in males; the median age in both sexes was 3 years, the mean age being 3 years in males and 3½ years in females. 8·2 per cent of all cases were manifest by 2½ years of age, 71·9 per cent by 3½, 88·0 per cent by 4½, 93·4 per cent by 5½ and 97·5 per cent by 6½.

(2) *The* 90 *per cent Manifestation Level.* As 87·6 per cent of the females and 97·0 per cent of the males manifested the disease by 4½ years old this age has been adopted, for the purpose of classification, as an arbitrary end-point at which about 90 per cent of the potential cases of scrapie may be expected; the 10 per cent developing after 4½ years of age are termed late manifestation. An affected animal is one alive and well at 4½ years old and not manifesting scrapie thereafter while under observation; an unaffected animal not under observation until 4½ years old is termed unclassified. An affected animal is one manifesting the disease at any age.

(3) *Sex Incidence.* The disease would appear to affect both sexes in similar proportions, although the data for males are few.

(4) *General Epidemiology.* The flock incidence, that is, the number affected per 100 breeding females per annum, varies widely from flock to flock, even of the same breed, and from year to year. Many flocks may be considered as free, that is, no manifest cases for at least 5 years; in many the incidence is low (1–3 per cent) or sporadic (< 1 per cent), in some it is medium (4–10 per cent) and in a few flocks it is high (> 10 per cent), and may exceed 20 per cent.

A group of flocks, each flock breeding all its own females and selecting most of its sires from flocks within the group, may be considered to be a closed breeding population or flock-group. The flock incidence varies widely between different flock-groups; several have had no cases, others many.

In an affected flock-group, many flocks remain free of the disease, although they have used sires reared in medium incidence flocks during a period when manifest cases were present.

The introduction into a free flock of a sire which afterwards manifested the disease while in the flock has not been followed by any further manifestation of scrapie, in one instance for 2½ years and in another for 1½ years thereafter.

Where sheep of another flock-group free of the disease are run on the same farm and intermingled with affected sheep, no case of scrapie has appeared in the free flocks over 3–5 years.

In recorded flocks with a medium to high incidence initially, the incidence has dropped to the low range in all but three flocks. In two of these it has not been possible to dispose of close relatives; in the third a low incidence has suddenly risen to the high level, coinciding with the female progeny of certain matings reaching the age of high manifestation.

Many flocks with a low or sporadic incidence initially have had no case for 2–3 years or longer.

We have found no evidence to suggest spread of the disease from affected animals to others of the same generation by contact, coitus or contaminated environment.

(5) *Familial Incidence.* While in individual flocks of low to medium incidence affected animals appeared to occur capriciously in different and unrelated families, when these data were studied with knowledge of the performance of the whole flock-group, it became apparent that the incidence among individuals with a close relative affected, that is, a full or half brother or sister, father, mother, aunt, uncle or grandparent, was very much higher than in individuals with no close relatives affected.

The incidence in a female age-group, that is, in the classified progeny born into one flock in one year, usually sired by 1 or 2 rams, is more variable than the flock incidence, and may exceed 60 per cent.

The incidence among the progeny of one affected parent is higher than the corresponding flock and age-group incidence; in free or sporadic incidence flocks it is usually 0–5 per cent, in medium incidence flocks 25–30 per cent, and in high incidence flocks 50 per cent.

An unaffected female producing affected progeny by one sire will usually produce unaffected progeny by other sires at previous and subsequent matings, and the pedigree of her descendants will show occasional affected individuals.

A pedigree derived from an animal with one affected parent will often show a generation unaffected, but the disease usually appears again within 2–3 generations.

The incidence does not appear to be influenced by order of birth or foster rearing of the progeny. The progeny born to mothers manifesting scrapie at the time of their gestation and suckling are often but not always affected; whether the incidence is significantly higher than might be expected is not known.

(6) *Heritability.* The manifestation of scrapie among 232 sheep coded according to the performance of their parents is shown in Table 1; the incidence observed follows closely that expected of a hereditary disorder controlled by an autosomal recessive gene.

Table 2 summarizes data for sheep with two affected parents, and for those sired by an affected ram in a flock which has had no other case of scrapie for at least 30 years, and in which the dams may therefore be presumed to be mostly homozygous 'clear'.

The occurrence of scrapie among 34 sets of gestational, probably binovular, twins is consistent with the data shown in Tables 1 and 2. One set born of 2 affected parents were both affected: of those born of an affected and an unaffected 'carrier' parent, sometimes both are affected, sometimes one and sometimes neither; while of 18 sets born of unaffected 'clear' parents, all were normal.

Of 49 progeny of affected sires and unaffected dams, 13 (26·2 per cent) were affected; of 5 progeny of

Table 1. MANIFESTATION OF SCRAPIE OBSERVED AMONG THE FEMALE OFFSPRING OF PARENTS WITH VARYING FAMILIAL CONNEXIONS WITH THE DISEASE, AND THAT EXPECTED FOR A SIMPLE RECESSIVE HEREDITARY DISORDER

Mating	Offspring No. at risk*	No. affected observed	 expected
Affected × affected	24	22	21·6
Affected × unaffected 'carrier'†	15	7	7·4
Unaffected 'carrier' × unaffected 'carrier'	6	0	~1 6
Unaffected 'carrier' × unaffected 'clear'‡	168	7	<8 2
Unaffected 'clear' × unaffected 'clear'	19	0	0

* Sum of affected plus unaffected animals under observation until the 90 per cent manifestation level at 4½ years old.

† Unaffected 'carrier' has one parent affected and/or more than one progeny affected.

‡ Unaffected 'clear' has no progeny or close relatives affected.

Table 2. MANIFESTATION OF SCRAPIE SHOWING THE CUMULATIVE TOTALS OF AFFECTED ANIMALS, OBSERVED AND EXPECTED ON THE BASIS OF A SIMPLE AUTOSOMAL RECESSIVE CHARACTER, AMONG THE OFFSPRING OF (*a*) TWO AFFECTED PARENTS, AND (*b*) ONE AFFECTED AND ONE UNAFFECTED 'CLEAR' PARENT

Age in years	½	1½	2½	3½	4½	5½	6½
(*a*) No. at risk*	93	63	42	31	30	29	28
Manifestation factor	0	0	0·08	0·72	0·88	0·93	0·97
No. affected							
Expected	0	0	3·4	22·3	26·4	26·1	27·1
Observed	0	0	5	23	25	27	27
(*b*) No. at risk*		36	27	12			
No. affected							
Expected		0	0	0			
Observed		0	0	0			

* Sum of affected plus unaffected or unclassified animals under observation up to this age.

unaffected 'carrier' sires and affected dams, 2 (40 per cent) were affected; while of 286 progeny of unaffected sires and unaffected dams, 24 (8·3 per cent) were affected.

The incidence among one age-group of 32 females sired by 3 rams in a flock with a very high incidence throughout the relevant 5 years was 34 per cent. Among the daughters of an affected ram it was 50 per cent, of an unaffected ram 14 per cent and of an unclassified ram 20 per cent.

(7) *Selection.* There is a special bias towards the selection of individuals which have made exceptionally rapid growth in infancy and adolescence with particular development of the skeletal musculature, for such animals tend to win the principal awards at our leading agricultural shows. Many 'show' winners of high excellence have close relatives affected, have affected progeny, or may even manifest the disease later.

(8) *Experimental Transmission.* 5 of 5 lambs inoculated intracerebrally with a brain suspension from a representative affected animal developed a scrapie-like disease 14–24 months thereafter; 5 controls showed no clinical abnormality.

These results are difficult to reconcile with the concept of an infectious disease of the usual communicable type.

The results lend strong support to the view that scrapie is primarily a hereditarily determined disease and establish that the clinical manifestation of the disease is highly heritable.

The results are consistent with the hypothesis that scrapie may be due to a single pair of alleles behaving in a simple Mendelian autosomal recessive manner. It is not possible to rule out other modes of inheritance, notably dominance with variable expression, or to assess the influence of modifying genes and of environmental factors upon manifestation.

As this hereditary scrapie can be transmitted by inoculation, a feature of scrapie amply demonstrated by others[4-6], it is suggested that in the homozygous recessive individual a substance, the transmissible scrapie agent, is formed which is closely associated with the development of the degenerative changes in several organ systems. Transmissible scrapie agent can be demonstrated in many organs[4,5], it is self-replicating[4,5] and its presence can be demonstrated in the experimental disease prior to the onset of clinical signs[5]. It is unusually stable to heat and chemical agents[5,6]. The immunological tests commonly present in virus infections are negative in scrapie[7].

It is postulated that scrapie is a hereditary disease of unusual type, in which the undesirable, indeed lethal, effects of the genotype are mediated through a positive factor, probably chemical in nature, which does not provoke the immune responses usual with self-replicating microbial infections. This concept would go far towards providing a satisfactory explanation of the facts at present known regarding scrapie. Further long-term studies will be necessary to provide adequate data to confirm the hypothesis, to elucidate the role of transmissible scrapie agent in the functional and anatomical pathology of the disease, and indeed to establish whether or not 'scrapie' represents more than one aetiological entity. A detailed report is being published elsewhere.

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[1] Wilson, D. R., *Vet. Rec.*, 64, 468 (1952).

[2] Parry, H. B., *Vet. Rec.*, 69, 43 (1957).

[3] Palmer, A. C , *Vet. Rev. Annot.*, 5, 1 (1959).

[4] Pattison, I. H., Gordon, W. S., and Millson, G. C., *J. Comp. Path.*, 69, 300 (1959).

[5] Stamp, J. T., Brotherston, J. G., Zlotnik, I., Mackay, J. M. K., and Smith, W., *J. Comp. Path.*, 69, 268 (1959).

[6] Gordon, W. S., *Vet. Rec.*, 58, 516 (1946).

[7] Chandler, R. L., *Vet. Rec.*, 71, 58 (1959).

THE PLEXIFORM ZONE AND SHAPE RECOGNITION IN THE OCTOPUS

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IT is the purpose of this article to suggest a possible structural basis for shape recognition in the octopus. The octopus is now the best-investigated animal in respect of its discrimination of visual shape, and this is chiefly due to the work of N. S. Sutherland. This author has also put forward a theory to account for his findings[1], but this theory was criticized by Dodwell[2] on grounds which seem to me to be substantially correct. The cephalopod visual system has also been studied from the neuroanatomical viewpoint by Kopsch[3], Lenhossek[4] and Cajal[5]. The present work is an attempt to infer the way the well-defined neural structures, uncovered chiefly by Cajal, result in the behavioural properties which the octopus displays when discriminating shape.

The System

The basis of the system is an arrangement which converts the distance between two points into an output, the size of which is inversely proportional to the distance. That is, the larger the distance between two imposed points the smaller will be the output, and the smaller the distance between the two points the larger the output.

This operation is performed by an arrangement of the following form. There is a set of lines running parallel to each other. Each line is interrupted at randomly chosen points, so that each line is divided into a number of segments of varying size.

Into this set of parallel segments enter at right angles a set of other lines (to be called retinal receptor fibres). These enter the layer of segmented lines at equally spaced intervals and penetrate the layer of the segmented lines to its other boundary.

Now let us assume that the retinal receptor fibres become active when they have been stimulated by a boundary discontinuity. The distance between two retinal receptor fibres reflects the distance between two points on a boundary. Let us also assume that the line segments in the parallel line layer also become active only when two or more than two simultaneously active retinal receptor fibres cross them. It will then follow that the closer together the two active retinal receptor fibres are, the greater the number of line segments will become active. The sum of active line segments in the parallel line layer will correspond in inverse ratio with the distance apart of the active incoming retinal receptor fibres.

The arrangement which has been described corresponds with what can be seen of the histology of the 'zona plexiforme' of the deep retina as described and illustrated by Cajal[6]. Fig. 1 shows the terminations of the incoming retinal fibres. Fig. 2 shows some of the fibres which may be identified with the segmented parallel line layer of our arrangement. It is to be seen that each of the segments in the section has an output leading out of the 'zona plexiforme' farther into the optic nervous system. We simply have to assume that the fibres in this figure act as a 'coincidence gate' or a spatial summator. If we make this assumption, then arrangements such as these have the function of converting distance between fibres into numbers of impulses. Distances or intervals can then be handled in the same way as intensities. Such a transducer of relational properties does not rely on 'scanning', with its high requirements of timing accuracy and overall precision. Such an arrangement would therefore have implications in the phenomena of neural integration beyond those of cephalopod vision.

So far, the arrangement described would transform the distance between a pair of points into an amount of neural activity The shapes to be discriminated, however, are composed of a great number of pairs of points. It is assumed that these pairs lie on lines parallel to each other and that each pair is measured by an arrangement such as was described above. This is done by having many such arrangements side by side, parallel to each other, running in the vertical visual axis. It is assumed that the total amount of excitation generated by the whole set of these arrangements gives the octopus its chief cue to the discrimination of shape. Different shapes are in this way translated into a single continuum of amount of activity. The problem of discriminating between them becomes one of differentiating between various levels of activity in a single channel.

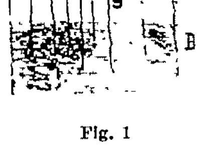

Fig. 1

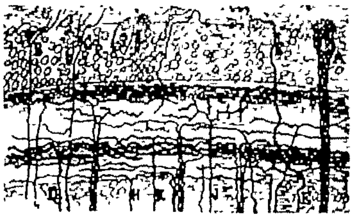

Fig. 2

Before proceeding, I should make clear that for the purposes of prediction two accessory assumptions must be made: (1) that vertical lines produce little or no output, because excitations on the same vertical arrangement inhibit each other over a small distance, and (2) that the octopus can also discriminate relative vertical positions of the output converted in the above way. This second appears to be a residual factor left when we have explained the rest of Sutherland's results. We may assume that it is an independent channel which signals these differences also as differences in quantity of output. It should also be stated that the present system has been left quantitatively flexible. It has been assumed, for example, that the vertical distance between two points leads to an output which is the reciprocal of this distance.

There is no reason for supposing that the conversion is precisely this. The theory only has to assume that the conversion is something like this, and then perhaps not over the whole range (for example, the system may collapse over very large visual angles).

Properties of the System

(1) *Transfer to different sizes of the same shape.* Let us take two parallel lines and their scale enlargement, and place them at right angles to the arrangements described above (that is, in the horizontal visual axis). The total amount of discharge from the system outlined above for the two figures will be the same. If we double the vertical distance between the lines, the amount of firing from each pair of points will halve. However, as we double the distance between the two lines, we also double their length. If we double their length we measure from twice the number of arrangements (as they run parallel to each other in the vertical direction). Therefore, the total amount of output given of both sets of lines separately will be the same. Transfer should therefore be excellent in the case of this shape.

Transfer from one size to another will depend on two factors: (*a*) the shape used; and (*b*) the other member of the pair from which the octopus has learnt to make the discrimination.

As regards the first point, it will be readily seen that, if we take a V shape lying on its side and a scale enlargement of it, the scale enlargement will produce a greater output. The small V shape is a part of the larger V shape. Such reasoning will apply to diamonds and triangles, *inter alia.* However, any scaling down (or scaling up) will subtract only the pairs of points (or add pairs of points) which are the most distant from each other on the vertical axis. These will be contributing disproportionately little to the total amount of output.

This brings me to the second point. If the total amount of output from the shape to be discriminated from the other member of the pair was larger, then the increase in output due to the enlargement of this shape will not destroy the discrimination, because the effective distance along the continuum is increased. (". . . the octopus, having been trained to discriminate between vertical and oblique, treats the horizontal in transfer tests as equivalent to the oblique (and vice versa for animals initially trained to discriminate between horizontal and oblique)"—Sutherland, 1958. p. 45*b*.) There may actually be an improvement if the other parameters which the octopus derives from the stimulus also increase the distance between discriminanda.

Sutherland has frequently noted that the octopus is capable of maintaining a discrimination between two shapes even when their size is altered (within certain limits) (for example, Sutherland, 1957*a*, p. 65, 1957*b*, p. 13). He has noted that a decrease in size shows a smaller transfer than an increase. This would be predicted on the present theory with certain pairs of figures where the distance along the continuum of output was decreased.

However, a breakdown of the size transfer data according to the shapes to be discriminated has not been considered. A search of Sutherland's data, however, reveals that there is a difference in the predicted direction (Sutherland, 1958). The animals were trained to discriminate between a square and a triangle, both 25 sq. cm. in area. Transfer tests to larger squares (36 sq. cm.) show 65 per cent correct responses. (The accuracy of responding to the original square under the same conditions is 65 per cent.) The figure for transfer to a small square is 67·5 per cent. However, transfer to larger triangles yields a score of 88 per cent in the case of the larger and 67·5 per cent when the triangle is made smaller (changes in area equal to those made in the case of the squares). As a triangle of the same area produces a much larger output than the square, an increase in the size of the triangle will increase the difference in the amount of output between the triangle and the square. Changes in the size of the square will not produce changes in output, if we assume (assumption 1) that its vertical lines produce little or no output. Transfer here should not vary with the direction of the change of size.

The theory is thus in agreement with what is known of the ability to transfer from one size to another in the octopus. Further testing of the theory should examine the relation between shape and the capacity to transfer to other sizes, and also the relation between size transfer and the other member of the pair to be discriminated. We should expect on this theory not only transfer of size to be relative to the other member of the stimuli to be discriminated. Even if the present theory is wrong, it will have been important if it directs attention to these two factors, which so far have been neglected.

(2) *Difficulty of discrimination between shapes.* The difficulty of discrimination on this theory should be proportionate to the difference in the quantity of output generated by the two shapes to be discriminated. Each pair of points on the boundary of the shape where it cuts a vertical arrangement will produce an output which will be inversely proportional to the distance of the two points. The sum of the outputs produced by all the pairs of points will characterize the shape for the octopus. The shape, regarding this mechanism only, will be represented as a quantity, or a number on a single dimension. The calculation of similarity can therefore be made with rigour. An index of similarity will be the difference between the two quantities generated by the two shapes to be discriminated. (Such a formulation assumes that the message on all other visual discrimination mechanisms is kept constant, such as that mentioned in assumption 2 (below). There are also others such as brightness, but these are easy to keep constant.)

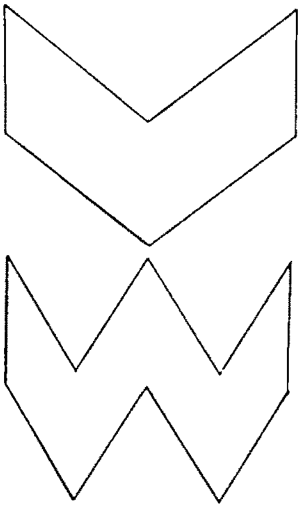

Fig. 3

(1) Sutherland[illegible] finds that horizontal rectangles are easily discriminated from vertical of the same proportions. This is readily explained. The horizontal rectangle stimulates a large number of vertical arrangements with a set of points relatively close to each other vertically. It is to be remembered that points close to each other produce a larger output than points farther apart. However, the vertical rectangle stimulates a relatively smaller number of vertical arrangements and by pairs of points farther from each other. There should, therefore, be a very large difference (1 : 25) between the outputs if the rectangles are 10 cm. × 2 cm.

"Perfect transfer occurred where the ratio of long to short sides was altered from 10 : 2 to 7 : 2 or 7 : 1" (Sutherland[1]). This would be an alteration at 1 : 25–4 : 49 and 1 : 49 in the amount of output in the discrimination. However, the absolute quantities of output would be different here also. As the direction of the ratios and of the difference remains the same here, this result does not count against the theory and, if anything, supports it. It is a result worth quoting because it shows that in circumstances where the differences among the two discriminanda are large, transfer occurs within a wide margin. It is as if the octopus was simply learning larger–smaller output differences. What would happen if larger numbers of shapes had to be retained in the same experiment by the octopus would be of interest. Further, these results show that if the two shapes to be discriminated give large differences of output, 'size-constancy' for the octopus does not need to be notably accurate.

(2) Sutherland[6] finds that two oblique rectangles (╱ and ╲) are indiscriminable. Here the sums of the outputs in the vertical arrangements are the same.

(3) Furthermore he stated[6]: "The finding that a vertical rectangle is more readily discriminable from an oblique than is a horizontal was unexpected". Such a difference is readily predicted from the present theory. The horizontal rectangle would produce a much greater output than the vertical rectangle. The output of the oblique rectangle would not be intermediate as might at first sight appear because the tilt of the figure would produce a set of points close to each other at the corners on the vertical axis. It is predicted that if the corners of the horizontal figure were altered to make it into a parallelogram with the short sides parallel to the vertical visual axis (the area being kept the same) the differential discrimination would disappear.

(4) Sutherland[7] has found that V and W shapes (Fig. 3) of the same height, width and area can be discriminated with great difficulty or not at all (accuracy 53 per cent) when the figures are in this orientation. An inability would be predicted on the present theory. The total vertical distances are the same in both cases. However, it would be predicted that the same shapes would be more discriminable when both are tilted through 90°. With ⩽ the vertical distance is divided into more short segments than in the case of <. Therefore, as short segments produce more output than long, and there are here more short segments, output should be larger for ⩽ than <.

(5) Sutherland[7] has found that ∨ is indiscriminable from ∧ (accuracy 50 per cent). This can be predicted in a straightforward way. He has also found that < can be discriminated from ∧ (though only with an accuracy of 56 per cent). The < shape would cut the vertical arrangements at points closer to each other than the ∧ shape, thus producing more excitation. However, ∧∧ is discriminated from > with greater ease (69 Per cent accuracy) and the present theory cannot readily account for this or the quantitatively small success of the octopus with < against ∧. (There are, however, assumptions about the sensitiveness to alterations in output with equal inaccuracies of verticality which differ for the shapes used and the amount of the shape deleted due to assumption 1 which could be made.)

(6) Sutherland (1958*a*) has found that octopuses have more difficulty in discriminating a diamond (a square with base at 45° to the horizontal) from a triangle than in discriminating a normal square from a triangle. This again is in accordance with the calculations of differences of magnitude of output derived from the theory. Though the other findings on the discriminability of the standard geometrical shapes (circles, triangles, etc.) show good agreement with the theory, differences in height (assumption 2) equally well account for some of the findings, as shapes of equal area were used. A further point to be borne in mind is that so far the assumption has usually been made that the placing of the shape on the receptor field of the octopus relative to the vertical has been geometrically exact, and departures from this have not been considered. However, the output of some shapes is much more affected by deviations. It is possible that the octopus uses this magnitude of oscillation about a mean to effect some discrimination. This supposition could be tested by imposing known and differing degrees of rotation in one direction and then the other in quick succession on the same shape, one amount serving as the positive signal and the other as the negative.

(7) It is found that the octopus can discriminate some up–down mirror images. This would be covered by assumption 2. However, left–right mirror images are indiscriminable, and this agrees with the theory.

Discussion

In the previous section, the salient reported findings on the shape discrimination of the octopus have been discussed in relation to the proposed system. The reader must be the judge of the success of the system in accounting for these findings. Predictions have also been made which allow the theory to be tested behaviourally.

However, the theory is not simply behavioural, and in this sense it is of much wider scope. A functional significance is suggested for a neuronal network of a kind to be found in many other places. It is proposed that this network converts relative distance into an output simply of quantity, and a simple *modus operandi* is given. In this way a look at the neurohistology may give a clue to the component operations on information in neural systems the overall function of which is already known. Direct work correlating visual input with events in the cephalopod optic lobes can also be used to check the correctness of my surmise.

On a more general plane it is conjectured that the octopus breaks down its visual information into various 'components' or dimensions of which the system put forward is only one. Each of these 'components' (given varying visual input) will vary only in amount. This amount for each 'component' is presumably represented as a quantity of excitation leaving the optic lobes, via one of the nerve fascicules. If this is correct, then sectioning one of these bundles, while conducting behavioural tests, would reveal the kind of classification or breakdown of the visual environment which the octopus performs by noting resulting confusions. Similarly, recording from these nerve bundles (should this prove feasible) while the eye is stimulated with various inputs should prove revealing. It is indeed possible that the transformation of shape into a simple quantitative continuum may prove to be a paradigm of the way that the problem of perceptual classification is dealt with in other organisms. In this way a given environment could be translated into a set of levels of activity in a number of channels. Thus a state of affairs for the organism could be represented as a point in an *n*-dimensional space.

My thanks are due to Prof. T. H. Bullock for discussion and encouragement, to Prof. J. Z. Young for helpful correspondence and neuroanatomical data, to Dr. N. S. Sutherland for discussion of his results, and to Dr. G. A. Horridge for helpful comment.

[1] Sutherland, N. S., *Brit. J. Psychol.*, 48, 55 (1957); *Nature*, 179, 11 (1957).

[2] Dodwell, P. C., *Nature*, 179, 1088 (1957).

[3] Kopsch, F., *Anat. Anzeiger*, 11, 361 (1896).

[4] Lenhossek, M., *Archiv Mikroskop. Anat. Entwicklungsgeschichte*, 47, 45 (1896).

[5] Cajal, S. R., *Trabajos del Laboratorio de investigaciones biologicas de la Universidad de Madrid*, 15, 1 (1917).

[6] Sutherland, N. S., *J. Comp. Physiol. Psychol.*, 51, 452 (1958); *Quart. J. Exp. Psychol.*, 10, 40 (1958).

[7] Sutherland, N. S., *J. Comp. Physiol. Psychol.*, 52, 135 (1959).

LETTERS TO THE EDITORS

GEOMORPHOLOGY

Bedrock Corrosion and Drainage Initiation by Seepage Moisture on a Gritstone Escarpment in Derbyshire

A SURVEY has been made of the podsolized soils, the morphology and the drainage of the areas known as Matlock Moor, Farley Moor, Tower Moor and Blackbrook Moor, which together form the larger part of the upper reaches or 'source basin' of the Bentley Brook, a tributary of the Derbyshire Derwent[1].

Observations have been made which show the corrosive action upon the underlying bedrock of water seeping through a 2-ft. deep layer of sandy colluvial material occupying a seepage hollow at the head of the brook. Such water, slow-moving, and with an appreciable content of humus, seeping vertically on to the bedrock, has corroded it to depths of 18–22 in. The layers below 18 in. in the whole profile are permanently wet and have been so even in the dry summer of 1959. The upper mineral layers of the profile are lacking in particles finer than 0 06 mm.; but it is not yet certain whether such particles are removed by normal vertical seepage to be redeposited at lower levels, or are removed by lateral movement of soil water, near the surface, in the wettest periods of the year.

Other observations provide evidence of the processes associated with the initiation of a semi-dendritic drainage pattern on a dip-slope, of less than 5°, adjacent to the hollow, and on the scarp slope to the west, overlooking the Sydnope valley. Integrated narrow seepage lines express themselves as overdeepenings of the solum upon such slopes, though there is no surface indication of their presence. A close grid of augerings across and along them shows significant changes of soil profile morphology both within the seepage lines and on the areas between them. These seepage lines link up with visible surface drainage at lower levels.

These two types of seepage action which affect the bedrock on the watershed between opposing dip and scarp streams do not suffice to break the crest of the escarpment. As a result of their headward extension, the seepage catchment of the watershed area becomes so small that the processes of corrosion and overdeepening can no longer continue except at some point downslope where enough moisture can assemble. Hence the crest is exhumed to form a rock platform on which tors may develop wherever the wide spacing of the master joints permits their survival.

The observation that a rock platform on the watershed between two opposing drainage systems may not only be preserved, but may actually broaden by exhumation, calls for a reappraisal of the current ideas on col-formation in the normal cycle of erosion, at least on uniformly hard rocks. It also leads to a hypothesis of tor formation applicable to some of the tors of the southern Pennines. The tors in question are formed during the gradual emergence of crestal rock platforms from adjacent bands of corroded bedrock and colluvial material. These successive bands form Penck's convex and intermediate slopes[2], although they are produced by seepage, not by drainage in visible channels.

The colluvium merges into a band of thin soils which then merges downslope into a mass of valley-head peat wherein the permanent source of integrated drainage is found. The peat often rests upon the rock identical with that of the tor. The rock is pierced in a pronounced knickpoint by the stream, which flows down over lower gritstone beds and intervening partings of shale before entering the wide main valley which is cut in the dominantly shaly rocks below the lowest grit and above the limestone.

There exists a possibility of assessing the period of time necessary to exhume these tor features. Most of them are 14–16 ft. in height. Young's estimate of surface lowering of the whole area of a small drainage basin, 17 miles to the north of the area considered here, is 0 005 in. per annum[3]. If one assumes a height of 15 ft. and that the tops of the tor features have not been lowered by weathering, then the time at which they began to be exhumed should be 36,000 years ago—a date which closely corresponds to the waning of the York glacier[4].

A more detailed account of this work will appear elsewhere. My thanks are due to Dr. G. Dury and Mr. R. A. G. Savigear for their comments and advice. This work has been supported in part by a grant from the Central Research Fund of the University of London.

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[1] Ordnance Survey, 1-inch map, 7th edit., Sheet 111, Buxton and Matlock (*SK*/3063).

[2] Penck, W., "Morphological Analysis of Landforms", 211 (Macmillan, 1953).

[3] Young, Anthony, *Proc. Yorks. Geol. Soc.*, 31, 2, 149 (1957).

[4] Flint, R. F., "Glacial and Pleistocene Geology", 395 (Wiley, New York, 1957).

GEOLOGY

Mid-Tertiary Stratigraphical Palæontology

IN the course of extensive economic palæontological researches based on world-wide investigations, it has been found necessary to review and re-assess the bases of mid-Tertiary correlation and dating. We confirm the validity of the five-fold subdivision of the Oligocene and Lower Miocene as represented by the faunas of the type localities of the Lattorfian, Rupelian, Chattian, Aquitanian and Burdigalian. These five faunal assemblages can readily be correlated through to Southern Europe, the Mediterranean, the Middle East, East Africa, Pakistan and India, and the Far East as a whole.

Although the classical type assemblages are mainly developed in a molluscan–echinoid facies we confirm that it is possible to correlate these facies with beds in Southern Europe, which also contain important and characteristic foraminiferal faunas which can readily be followed through to the Far East, notwithstanding that minor stratigraphical and taxonomic revisions of older work are necessary.

In brief, beds equivalent to the Lattorfian are characterized by *Nummulites intermedius* and *N. vascus* without *Eulepidina*; the equivalents of the Rupelian are characterized by the persistence of *Nummulites intermedius* and *N. vascus* and the appearance of *Eulepidina*; the equivalents of the Chattian (where confirmed) are characterized by the persistence of *Eulepidina* and other Rupelian Foraminifera, but all true *Nummulites* had become extinct. Following the mid-Tertiary stratigraphical break, the equivalents of the Aquitanian (forming the lowest beds of the Miocene) are characterized by the persistence of *Eulepidina, Spiroclypeus* and *Miogypsinella complanata,* and the incoming of many forms, such as *Miogypsina* s. str., *Meandropsina anahensis* and *Miolepidocyclina,* which constitute part of the Neogene evolutionary surge; the Burdigalian equivalents are characterized by such forms as *Ostrea latimarginata, Borelis melo curdica, Flosculinella bontangensis* and *Taberina malabarica,* by the persistence of *Miogypsina* and *Austrotrillina howchini,* and by the appearance of *Borelis melo,* but *Eulepidina* and *Spiroclypeus* had become extinct.

It can be evaluated from the above that in the Far East the '*c*' stage is probably Lattorfian, the '*d*' stage is Rupelian, the '*e*' stage is Aquitanian, and the 'f_{1-2}' substages are Burdigalian; the presence of the Chattian is not proved. Besides the fossils mentioned above there are many others which substantiate the above conclusions.

Within the equivalents of the five classical stages there is a succession of planktonic foraminiferal faunas (Globigerinaceae) which appears to be constant wherever found and studied. These planktonic successions can be firmly dated in terms of the classical stages by their associated faunas; they permit accurate age determination of marine faunas in the western hemisphere, where, because of relative isolation, few benthonic fossils are in common with their Eurasian and African equivalents.

It has been possible to demonstrate the equivalence of the Caribbean zones of *Globorotalia fohsi fohsi, G. fohsi lobata* and *G. fohsi robusta* to the Burdigalian. The Aquitanian seems to be equivalent to all the planktonic foraminiferal zones erected by Bolli (1957) for the Cipero formation of Trinidad below the *G. fohsi fohsi* zone; consequently, all the Vicksburgian and its equivalents are Aquitanian. The evolution of *Orbulina,* known to occur in the upper part of the *Globigerinatella insueta* zone of the Caribbean, also occurs in the highest Aquitanian of the Mediterranean and Australia; this correlates with the first appearance of *Orbulina* in the upper '*e*' stage of the Far East.

It seems to be very important to note that, on the basis of the succession of planktonic foraminiferal faunas, no Oligocene can be recognized in the marine sediments of the western hemisphere, as far as published records show. This absence of Oligocene has now been confirmed by the discovery of a planktonic foraminiferal fauna associated with true Oligocene larger foraminifera such as *Nummulites intermedius* and *N. vascus* in East Africa. This planktonic foraminiferal fauna is being described elsewhere; it is noted here as having common elements with similar faunas seen in the Rupelian of south Germany and that described by Russian workers. This fauna has never been recorded or observed by us in Central American areas, and is missing precisely where we place the major stratigraphical break in that region.

The absence of marine Oligocene in the Central American region is again confirmed by the presence of *Miogypsina* s. str. associated with *Eulepidina* and of *Pliolepidina* in the lowest post-Eocene of the well-known southern Trinidad succession, which contains the oldest known post-Eocene planktonic zone of the Caribbean. True *Nummulites* (as distinct from *Palæonummulites*) is absent above the Eocene in the Caribbean.

Palæonummulites Schubert 1908 (type species *Nummulina pristina* Brady 1874) is regarded as a prior synonym of both *Operculinella* Yabe 1918 and *Operculinoides* Hanzawa 1935. The lectotype of *Palæonummulites pristina* (Brady) has been isolated and it will be described elsewhere, together with an emendation of the genus.

The genus *Pliolepidina* H. Douvillé 1915 is now regarded as a prior synonym of *Multicyclina* Cushman 1918, *Multilepidina* Hanzawa 1932, *Cyclolepidina* Whipple 1934, and *Pliorbitoina* Van de Geyn and Van der Vlerk 1935. It is believed that all species referable to this genus, whether originally recorded from the Far East, the Mediterranean region, East Africa, or Central America, are of Miocene age. The large and significant unconformity present in the Central American region is marked by the presence of much re-working of material into the basal Miocene and this has given rise to much stratigraphical confusion as to the age of the beds in question. Indeed, many of the lowest Miocene beds present have been dated as Eocene, purely on the presence of re-worked Eocene fossils, but the occurrence of *Pliolepidina* often shows that they are of lowest Neogene age; examples are the San Fernando formation of Trinidad and part, at least, of the Gatuncillo formation of Panama.

Full details of our work will be published in the near future elsewhere, together with a full bibliography.

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Oriented Quartz Inclusions in Garnets

METAMORPHIC garnets in the Central Highlands of Scotland frequently contain oriented inclusions of quartz, which throw light on the mechanism of deformation of the latter mineral. In particular, garnets which have an S-shape arrangement of inclusions and have consequently grown at the same time as deformation show a variety of morphogenetic types of quartz grains, which were incorporated into garnet as inclusions.

In a specimen of mica-amphibole-garnet schist from the River Tummel (National Grid No. *NN.* 899605) there are three types of such grains. In the centre of the garnet there is a number of crescentic inclusions (about 0·05 mm. × 0·01 mm. in size), which have no visible strain shadows, glide lamellæ or deformation bands, but which in cross-polarized light are nevertheless optically inhomogeneous, although even at high magnifications the constituent units responsible for the inhomogeneity cannot be recognized. Associated with these grains, but continuing towards the periphery of the garnet, there are much larger elongated quartz inclusions (about 0·15 mm. × 0·1 mm. in size) which are optically homogeneous and are not crescentic in shape. Lastly, at the periphery of the garnet there are relatively equidimensional grains, again much larger than the first variety (0·1–0·15 mm. across), and which may have strain shadows when the grains are continuous into the matrix.

In the crescentic inclusions the average optic axis of the constituent quartz is normally slightly oblique to the elongation of the inclusions, while in the elongated optically homogeneous variety the optic axis tends to be parallel to the elongation of the grain. In the peripheral grains no constant relationship can be established between the position of the optic axes and the dimensions of the inclusions. The S-shape trend of inclusions implies that the innermost grains have been included into the garnet early in the process of movement, while the outermost individuals record the immediately post-movement stage of lattice re-organization. It is clear that throughout the deformation the fabric of the rock coarsened progressively and that the early dimensionally oriented fabric is conditioned by movement, whereas the later fabric did not develop dimensional orientation, and was presumably formed under static conditions. Since at an earlier stage the sites now occupied by the dimensionally unorientated fabric must have had quartz grains like those preserved in the interior of the garnet, the peripheral quartz has suffered severe re-organization.

Fellows[1] recognized that deformed crystalline quartz undergoes first recrystallization and then neo-crystallization. In the garnet described, the crescentic inclusions reflect the initial deformation, while the elongated optically homogeneous grains have recrystallized. These grains are analogous to mimetic mica flakes, and those in the matrix, according to this interpretation, are neo-crystalline. Both recrystallization and neo-crystallization are recognized in metallurgy by Burke and Turnbull[2]. The deformed crescentic grains can be interpreted as polygonized grains (cf. Cahn[3]) retaining their initial fine deformation texture, which is not resolved under a microscope. On the other hand, the strain-shadows in the peripheral grains must have originated during an episode of renewed deformation which succeeded the neo-crystallization.

The fact that there are slight differences—in the relationships of the optical orientation and elongation—between the crescentic (polygonized) and recrystallized grains suggests that recrystallization affects the overall lattice orientation, and that this factor must be considered when the behaviour of metamorphic quartz is studied.

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[1] Fellows, R. E., *Bull. Geol. Soc. Amer.*, 54, 1399 (1943).

[2] Burke, J. E., and Turnbull, D., "Prog. Metal Phys", 3, 282 (1952).

[3] Cahn, A. W., *J. Inst. Metals*, 76, 121 (1949).

PHYSICS

A Theory of Ball Lightning Formation

In a paper which has recently attracted a considerable amount of attention, Kapitza[1] has proposed a standing electromagnetic wave description of ball lightning. His paper presents a good qualitative explanation of this effect in terms of the resonance absorption of a standing wave due to a conducting ionized sphere of plasma, occurring when the wavelength of the radiation is approximately four times the diameter of the fireball.

It is the purpose of this communication to point out that a polarized electromagnetic standing wave can actually produce containment of charged particles in the neighbourhood of its electric nodes, and that this effect should be important in the initial stages of fireball formation. As a result of this investigation it has been shown that it is possible for electrons to acquire sufficient energy in the electric node regions to produce further ionization by collision, provided that the standing electromagnetic wave yield-strength is in the vicinity of 10^6 V./m. At first sight this figure may seem unusually high; nevertheless it should be recalled that field strengths of this magnitude are already required to cause atmospheric breakdown during thunderstorms. It is proposed, therefore, that the atmosphere is initially very weakly ionized (due to the thunderstorm) and that the regions around the electric nodes rapidly become highly ionized due to electron ionization by collision, building up in an avalanche fashion until the plasma frequeney ω_p is greater than the frequency of the applied field ω. At this point further field penetration of plasma becomes impossible, so that surface absorption of energy becomes responsible for maintaining the fireball in a manner similar to that suggested by Kapitza. Moreover, the spherical shape of fireballs fits into the ultimate surface absorption picture quite well, since this is a direct consequence of the fact that the configuration of minimum surface energy requires a spherical distribution.

The actual containment of a charged particle in a polarized standing electromagnetic wave will be demonstrated using a straightforward technique similar to that developed by R. Wuerker (unpublished private communication) and Cushing and Sodha[2].

We shall assume that a polarized standing electromagnetic wave can be represented by

$$B = B_0 \sin \omega t \cos \frac{2\pi x}{\lambda} \hat{k} \qquad (1)$$

$$E = -cB_0 \cos \omega t \sin \frac{2\pi x}{\lambda} \hat{\jmath} \qquad (2)$$

where B_0 represents the maximum value of oscillating field of frequency ω, and wave-length λ, and $\hat{\imath}$, $\hat{\jmath}$, $\hat{k}$ are the usual cartesian unit vectors.

The following equations of motion of a charged particle may be derived using equations (1) and (2) and the Lorentz force equation:

$$\ddot{x} = \frac{eB_0}{m} \dot{y} \sin \omega t \cos \frac{2\pi x}{\lambda} \qquad (3)$$

$$\ddot{y} = -c\frac{eB_0}{m} \cos \omega t \sin \frac{2\pi x}{\lambda} - \frac{eB_0}{m} \dot{x} \sin \omega t \cos \frac{2\pi x}{\lambda} \qquad (4)$$

$$\dot{z} = 0 \qquad (5$$

Integrating equation (4) we immediately obtain :

$$\dot{y} = -\frac{c}{\omega}\frac{eB_0}{m}\sin\omega t\sin\frac{2\pi x}{\lambda} + C \qquad (6)$$

where the constant C represents the initial velocity of the particle in the y-direction at time $t = 0$.

Direct substitution of equation (6) into (3) gives :

$$\ddot{x} + \frac{c\omega}{4}\left(\frac{eB_0}{m\omega}\right)^2\left\{1 - \cos 2\omega t\right\}\left\{\sin\frac{4\pi x}{\lambda}\right\} = \frac{eB_0}{m}C\sin\omega t\cos\frac{2\pi x}{\lambda} \qquad (7)$$

Equation (7) is clearly a difficult equation to solve ; however, if we concern ourselves with solutions of (7) representing oscillations of small amplitude about the electric field nodes, we may write :

$$\cos\frac{2\pi x}{\lambda} \simeq 1 \qquad (8a)$$

$$\sin\frac{4\pi x}{\lambda} \simeq \frac{4\pi x}{\lambda} \qquad (8b)$$

and equation (7) can be reduced to the form :

$$\ddot{x} + \frac{1}{2}\left(\frac{eB_0}{m}\right)^2\left\{1 - \cos 2\omega t\right\}x = \frac{eB_0}{m}C\sin\omega t \qquad (9)$$

which is in the form of a driven Mathieu equation. The solutions of the Mathieu equation have bound or unbound characteristics depending upon the choice of parameters[3]. For example, when :

$$\frac{1}{2}\left(\frac{eB_0}{m\omega}\right)^2 < 0{\cdot}65 \qquad (10)$$

the system operates in the first region of stability, and the solution of equation (9) takes the following bound form :

$$x = F\left\{\cos\omega_0 t - \frac{a}{8}(1 + \sqrt{a})\cos(2\omega - \omega_0)t - \frac{a}{8}(1 - \sqrt{a})\cos(2\omega + \omega_0)t + \ldots\right\} + G\left\{\sin\omega_0 t - \frac{a}{8}(1 + \sqrt{a})\sin(2\omega - \omega_0)t - \frac{a}{8}(1 - \sqrt{a})\sin(2\omega + \omega_0)t + \ldots\right\} \qquad (11)$$

where F and G are constants of integration and ω_0 represents the oscillating frequency of the particle about the electric node along the x-axis. Where we have defined :

$$a = \frac{1}{2}\left(\frac{eB_0}{m\omega}\right)^2 \qquad (12)$$

and

$$\omega_0 = \frac{eB_0}{\sqrt{2}m}\sqrt{1 + \left(\frac{eB_0}{4m\omega_0}\right)^2} \qquad (13)$$

It is convenient to represent the harmonic binding of the particle about the node by means of an effective potential. In the first region of stability this assumes the form :

$$V(x) = \frac{1}{2}\left(\frac{m}{e}\right)\omega_0{}^2x^2 \text{ volts} \qquad (14)$$

By the substitution of condition (10) into equation (6) it is possible to find the y-motion about the electric nodes, and it has been shown that the charged particle is quasi-bound in this direction. The particle will describe Lissajous's figures in the xy plane, with dominant frequencies of ω_0 and $(\omega-\omega_0)$ in the x and y directions, respectively. Having convinced ourselves that a charged particle can be bound in the vicinity of an electric node, it will suffice to say that a corresponding investigation of solutions in the neighbourhood of the electric anti-nodes yields no bound solutions ; in fact, indicates that the particles are pushed away from the anti-nodes toward the nodes. Although a charged particle is essentially contained in the x–y plane, it is clear that there is no binding in the z-direction. Nevertheless the x–y binding will allow the particle to be 'contained' in such a manner as to allow it to produce considerably more ionization by collision before drifting out along the z-axis, ultimately leading to a region of high plasma density and conductivity in the electric node neighbourhood. On the basis of Kapitza's arguments, standing wave frequencies in the neighbourhood of 10^9 cm./sec. would be required to explain the 'observed' fireball sizes of 10–20 cm. Putting this value of ω into condition (10) along with the required electric field strength of 10^6 V./m., it can be seen that the condition for containment in the first region is easily satisfied. In addition, Kapitza has suggested that a fireball is located at the electric field anti-node, whereas our description indicates that the formation process would be expected to take place at the electric node.

Finally, it is suggested that this theory could be tested under laboratory conditions. This could be accomplished by using a standing wave created by a pair of travelling waves fed from opposite sides of the containment region. This is necessary in order to preserve the symmetry about the y–z plane, after the plasma conductivity has reached a point where surface absorption is predominant. Theoretical investigation of genuine three-dimensional plasma containment is at present in progress using three separate standing waves parallel to the x, y and z axes, respectively, the results of which will be published later elsewhere.

I would like to acknowledge many useful and illuminating discussions with Mr. B. Bloch during the initial period of this investigation, and with Drs. R. Wuerker and H. C. Corben during the latter part.

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[1] Kapitza, P. L., *Doklady, U.S.S.R.* (1955).

[2] Cushing, V., and Sodha, M. S., "Confinement of Plasma by Standing E-M Waves", "Physics of Fluids" (to be published).

[3] McLachlin, N. W., "Theory and Applications of Mathieu Functions" (Oxf. Univ. Press, New York, 1947).

Neutron Diffraction Study of Antiferromagnetic Spin Waves in α-Ferric Oxide

IT has been shown by Elliott and Lowde[1] that spin waves will manifest themselves in the neutron scattering by a crystal as coherent diffuse peaks near the elastic reflexions. In particular, the dispersion relation of the spin waves may be directly obtained from the way the diffuse peak-width Γ changes with $\theta - \theta_B$, the angular missetting of the crystal from the Bragg setting. Such measurements have been done on crystals of iron[2] and magnetite[3], and in both cases a quadratic dispersion law was found.

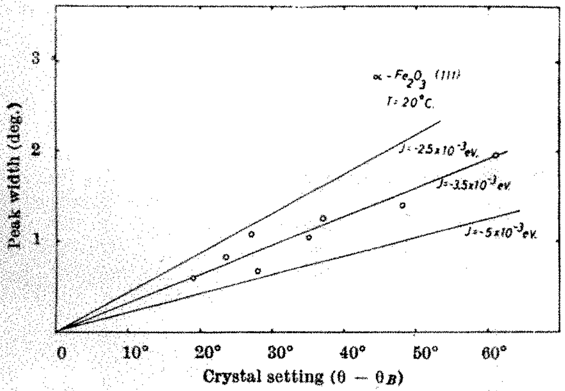

Fig. 1. Diffuse peak width plotted versus angular missetting from Bragg position for (111) reflexion. Straight lines have been calculated for different spin-wave velocities, corresponding to the indicated values of the predominant exchange integral

It may be shown on general grounds that in an antiferromagnet the dispersion law for large wavelengths is linear, so that there is a magnon propagation velocity c_m independent of frequency. As a consequence the relation between Γ and $\theta - \theta_B$ must also be expected to be linear.

Measurements of diffuse peak widths of antiferromagnetic α-Fe_2O_3 (hematite) near the (111) reflexion (rhombohedral indices) indeed show this linear dependence (Fig. 1). From the slope a spin-wave velocity $c_m = 3{\cdot}76 \times 10^6$ cm./sec. is deduced.

A theoretical expression for the spin-wave velocity may be found by solving the secular equation arising from a Heisenberg-type exchange coupling between the spins of the iron atoms, which in hematite belong to four different sub-lattices. The expression contains atomic position co-ordinates and the exchange integrals. The latter have been discussed by Gilleo[4] in terms of super-exchange interactions between iron spins through intervening oxygen atoms. These interactions turn out to be of four types, differing in interatomic distances and angles, and with different values of the exchange integral J. If now, following Gilleo, one ignores all interactions except those belonging to the type with the largest $|J|$, the expression for the spin-wave velocity becomes:

$$c_m = -2^{5/2}\, JSa/\hbar \qquad (1)$$

where S is the iron spin and $a = 5{\cdot}03$ Å. the base of the hexagonal unit cell. The value of c_m is actually slightly dependent on direction of propagation, but this anisotropy is within 4 per cent. Substituting the observed c_m in (1) one finds for the predominant exchange integral $J = -0{\cdot}0035$ eV., the minus sign indicating an antiferromagnetic coupling.

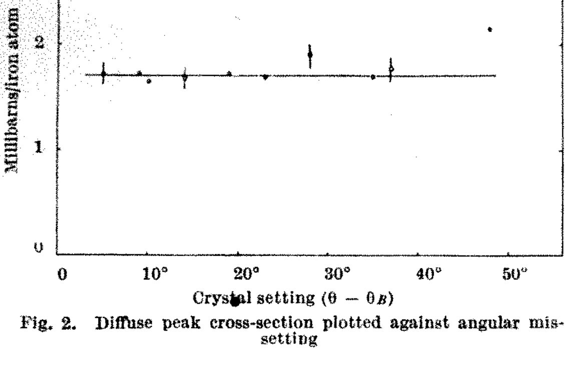

Fig. 2. Diffuse peak cross-section plotted against angular missetting

The value of the exchange integral thus found may be used in a calculation of the diffuse scattering cross-section. Elliott and Lowde[1] have given an expression for the cross-section integrated over the whole diffuse peak for a simple class of antiferromagnets and shown that it is independent of crystal missetting. Their treatment may be generalized to cases including that of hematite for which, with the above value of the exchange integral, it leads to a constant cross-section of 1·4 millibarns per iron atom. The observed values are plotted in Fig. 2 for different angles of misset and show this general behaviour, although with a somewhat higher cross-section value of 1·7 millibarns.

It remains to prove that the observed diffuse peaks are indeed due to antiferromagnetic spin waves and not to lattice vibrations, which also would show a linear dispersion relation. Since the nuclear structure factor of the (111) reflexion is zero, such vibrations could only manifest themselves as magnetic vibrational scattering. To distinguish between the two kinds of scattering one may use their different dependence on the direction of spin alignment. The spin wave cross-section contains a factor $(1 + e_z^2)$, where e_z is the cosine of the angle between the spin direction and the normal to the reflecting plane. The magnetic vibrational scattering, on the other hand, is proportional to $(1 - e_z^2)$, as is the elastic magnetic scattering. This effect has been used in the study of ferromagnetic spin waves, where e_z may easily be varied by an external magnetic field.

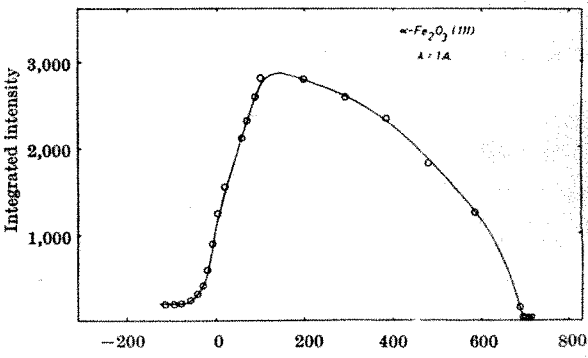

Fig. 3. Temperature variation of the antiferromagnetic (111) Bragg reflexion

In the case of antiferromagnetic spin waves this possibility does not exist. However, in hematite the spin direction is dependent on temperature. Fig. 3 shows the temperature variation of the elastic scattering in the (111) Bragg peak. It is in agreement with the observation of Shull *et al.*[5] that at low temperatures the magnetic moments are pointing along the [111]-direction ($e_z = 1$), whereas at high temperature they are in the (111)-plane ($e_z = 0$). From Fig. 3 and the fact that the inelastic scattering is proportional to absolute temperature, it may be inferred that when the crystal is cooled from 20° C. to − 35° C. the intensities due to spin waves and lattice vibrations should change by + 12 per cent and − 92 per cent respectively. Experimentally an increase of 10 ± 5 per cent was obtained, showing that the diffuse peaks must be of almost pure spin-wave origin.

A complete account of this work will be published elsewhere.

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[1] Elliott, R. J., and Lowde, R. D., *Proc. Roy. Soc.*, A, **230**, 46 (1955).
[2] Lowde, R. D., *Proc. Roy. Soc.*, A, **221**, 206 (1954).
[3] Riste, T., Blinowski, K., and Janik, J., *J. Phys. Chem. Solids*, **9**, 153 (1959).
[4] Gilleo, M. A., *Phys. Rev.*, **109**, 777 (1958).
[5] Shull, C. G., Strauser, W. A., and Wollan, E. O., *Phys. Rev.*, **83**, 333 (1951).

Experiments with Plasma Rings

The use of a coaxial plasma gun with radial magnetic field for the production of magnetized plasma rings was suggested[1] by H. Alfvén in 1958. Experiments with a plasma gun of that type were started in the spring of 1958 and a detailed report is to be published[2]. We present here some new results obtained during continued experiments with a slightly modified plasma gun.

A detailed description of the equipment is given in the previous paper[2]. The main features are described here with reference to Fig. 1, which shows a cross-section of the apparatus. Between the coaxial electrodes of the plasma gun a discharge is set up in hydrogen with a pressure in the range of 0·03–3 torr. The energy, initially stored in the condenser bank of 200 μF., charged to 5–8 kV., is released when the spark gap is triggered by a high-voltage pulse. The electrodynamic force in the plasma pushes the ionized gas towards the muzzle of the gun. Here it encounters a static magnetic field, which is directed radially. The field is maintained by an electromagnet. The central conductor of the gun, which is made of iron, serves as north pole. The outer conductor is of stainless steel, except for the muzzle where an iron plate forms the south pole of the magnet.

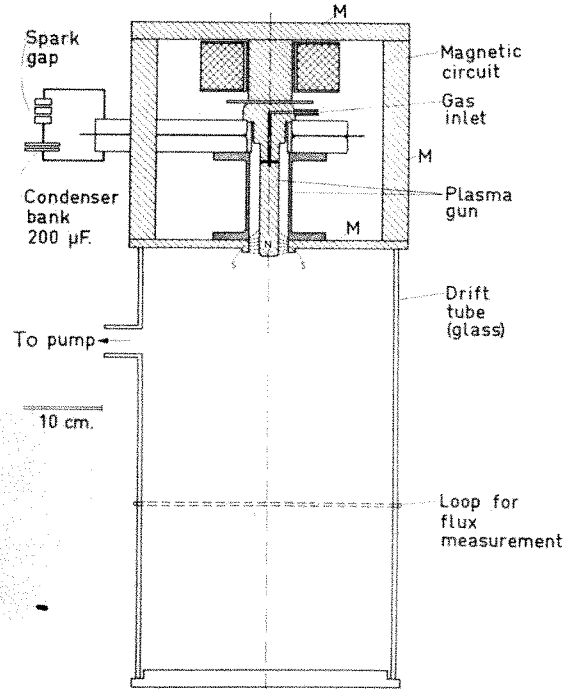

Fig. 1. Cross-section of plasma gun with drift tube

When the accelerated plasma reaches the muzzle it has a high conductivity, so that the magnetic lines of force cannot penetrate it. They are therefore pushed in front of the plasma, down into a drift tube of glass, which is located under the gun. Here a rapid expansion of the plasma towards the glass wall of the tube takes place and a ring of magnetized plasma is formed, through which the stretched field lines of the static field close.

The plasma is studied by means of high-speed photography, using Kerr-cell cameras, and by magnetic measurements. The magnetic flux carried by the plasma is measured by loops around the glass tube connected to integrating resistance-capacity circuits ($R = 10$ k.ohm, $C = 0{\cdot}05$ μF.).

Fig. 2 shows a typical picture of the discharge taken 4 μsec. after the ignition. The luminous front, which probably is due to a shock wave preceding the magnetized plasma, reaches the bottom of the drift tube at about 9 μsec.

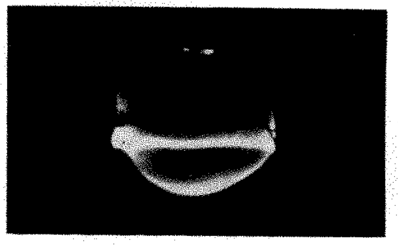

Fig. 2. Kerr-cell photography of the discharge taken 4 μsec. after the ignition

Fig. 3 shows oscillograms of the circuit current and the magnetic flux measured by a loop situated 40 cm. below the muzzle of the gun. The circuit current performs a strongly damped oscillation with a peak value in the first half-cycle of 220 k.amp. and passes through zero 12 μsec. after the ignition. The magnetic flux increases to a maximum, which often appears when the circuit current passes through zero, and then decays approximately exponentially. As the circuit current passes through zero the flux curve is smooth and the direction of the flux through the loop does not change. This indicates that a current continues to circulate in the plasma independently of the circuit current, which is rapidly damped out. Even at 40 μsec. after ignition, a toroidal current is circulating in the plasma.

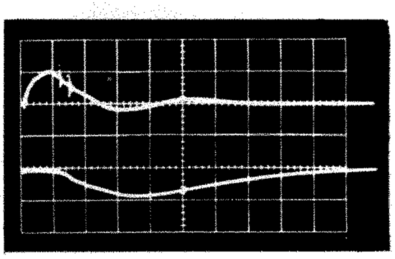

Fig. 3. Oscillogram of the main circuit current (upper curve) and the magnetic flux measured by a loop around the drift tube 40 cm. below the muzzle of the gun and recorded after integration. Time-scale, 5 μsec./div., current scale, 220 k.amp./div. Flux scale, 1 mWb./div. The applied static flux in the plasma gun was 0·47 mWb.

One would expect that the ring-shaped plasma passing the static magnetic field at the muzzle of the gun would conserve the magnetic flux through the central conductor of the gun as a 'poloidal' flux through the hole of the plasma ring formed. Often, however, the flux carried by the plasma is several times greater than the static flux. Whether this can be ascribed to asymmetry in the discharge or whether it depends on some mechanism by which the toroidal flux in the ring is transformed to a 'poloidal' flux is not clear.

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[1] Alfvén, H., Proc. Second Int. Conf. on Peaceful Uses of Atomic Energy, Geneva, **31**, *P*/145 (United Nations, New York, 1958).

[2] Alfvén, H., Lindberg, L., and Mitlid, P., *J. Nuclear Energy*, Part C (in the press).

[3] Dattner, A., Proc. Fourth Int. Conf. on Ionization Phenomena in Gases (Uppsala, 1959).

CRYSTALLOGRAPHY

Spiral Growth of Colloidal Gold and Moiré Fringe

MANY examples of spiral growth have been reported since Frank[1] introduced the idea of screw dislocation into the general mechanism of crystal growth. Amelinckx[2] has reported growth spirals which appeared on chemically deposited crystals of gold. We also wish to report various growth spirals and related matters recently observed by electron microscopy on lamellar single crystals of gold, prepared colloid-chemically under definite conditions as discussed in previous papers[3]. Brüche and Demny[4] also reported similar patterns on gold crystals and discussed the growth mechanism.

Fig. 1*A* shows a typical example of a lamellar single crystal of colloidal gold having the growth spiral. A small hole exists at the centre of the spiral, from where we can follow the ridge line and, after passing on the outer perimeter of the crystal, we can come back to the initial starting point. The three-dimensional configuration of such a crystal is suggested by Fig. 1*C*, which is an electron micrograph of a specimen replicated by carbon film *in vacuo*, to be in a manner demonstrated by Fig. 1*G*, though slightly exaggerated in its step height. The two spirals on both surfaces of a crystal are nearly symmetrical to each other in most of the present examples. This seems very reasonable as the specimens were grown in a suspension and the condition of the medium was quite the same on both sides of each crystal during growth. Fig. 1*B* shows another example where a rather regular hexagonal loop appeared. We can also observe a small hole as well as the returning loop as indicated by arrows.

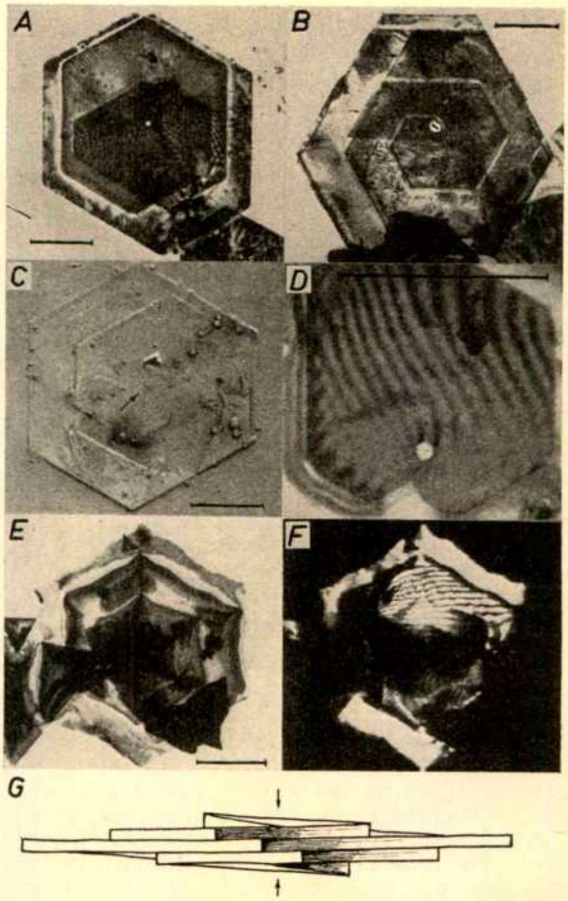

Fig. 1. Electron micrographs and schematic demonstration of single crystals of colloidal gold with growth spirals. Each scale gives 1μ. In Fig. 1*G*, the two arrows show the position of a core of screw dislocations

One of the most outstanding features in the present specimens is that sharp moiré fringes appeared on the growth steps when observed under an electron microscope. Various causes for the moiré fringe have been reported so far[5]. For the present case, we think it reasonable that it is mainly caused by the slight azimuthal rotation of each step, which takes place against another step on which the former one lies and around an axis parallel to the Burgers vector of the screw dislocation, which in turn is normal to the flaky (111) plane of the crystal. Neither considerable separation nor relative inclination of these steps was detected so far as the methods of shadow casting and carbon replica were concerned. We have also taken a set of dark-field images of moiré fringes formed with the electron beam reflected by the lattice plane contained in the region of the crystal where two or more steps were superposed. Fig. 1*F* shows an example of these dark-field images formed with the $(2\bar{2}0)$ reflexion of the crystal, the ordinary electron micrograph of which is shown in Fig. 1*E*. In the dark-field image, there appeared nearly parallel thick moiré fringes, the direction of which is again parallel to $[2\bar{2}0]$ axis of the crystal. This means that they were formed by the interaction of two electron beams reflected respectively by each $(2\bar{2}0)$ plane contained in two superposed steps. The moiré fringe is certainly very sensitive to the slight distortion of the crystal. But when the cause of the fringe is supposed to be attributable only to the slight azimuthal rotation of steps, the angle of rotation α is given by an equation[6]: $\alpha = d_{2\bar{2}0}/D$, where D and $d_{2\bar{2}0}$ are the spacing of the moiré fringe and the interplanar spacing of the $(2\bar{2}0)$ plane. With the set of dark-field images, the angle α has been estimated to be of the order of 2×10^{-3} rad. on an average. Further, it increases roughly linearly from 7×10^{-4} rad. to 4×10^{-3} rad. or to much larger values as the position on the step, where the spacing of the fringe was measured, approaches the centre of the crystal along the spiral step. The application of the same method to another specimen also gave results of the same order for the angle of rotational misorientation.

As for the small hole, we found it on some 60 of about 100 crystals with the spiral marking. Further, it can also be observed in Fig. 1*C* as marked by an arrow, where the original crystal has not suffered from any damage by the electron bombardment. Fig. 1*D* is an enlarged photograph of a hole which is a nearly regular hexagon in shape. Its edge line is oriented in parallel to each ridge line of the step. The growth spiral starts from one apex of the hexagon with round ridge line at the outset. Frank[7] has derived the relationship between the hole diameter (D_0) and the strength of the Burgers vector (b) at the core of the screw dislocation as follows, though slightly modified here:

$$b = 2\pi(S \cdot D_0/G)^{\frac{1}{2}} \quad (1)$$

where S and G are the surface free energy and modulus of rigidity of the material. The hole diameter is distributed over a range of 200–2400 A. and the most probable value is concentrated in a range of 800–1200 A., while the mean value becomes 1020 A. With this value, the strength of the Burgers vector was estimated to be 93–118 A., where $2\cdot76 \times 10^{11}$ dyne/cm.[2] and 600–1,000 erg/cm.[2] were adopted for G and S[8] respectively. On the other hand, we have also estimated the step height to be 109 A. on an average by the shadow-casting method. This is of quite the same order as the strength of Burgers vector above obtained. Thus, if the step height is supposed to correspond directly to the Burgers vector, Frank's theory has been well proved in the present case.

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[1] Frank, F. C., *Disc. Farad. Soc.*, **5** 48 (1949).

[2] Amelinckx, S., *Phil. Mag.*, **43**, 562 (1952).

[3] Suito, E., and Uyeda, N., Proc. Internat. Conf. E.M., 224 (London, 1954); Vierter Internat. Kongr. Elektronenmikros. (in the press).

[4] Brüche, E., and Demny, J., *Naturwiss.*, **46** 225 (1959).

[5] Menter, J. W., "Advance in Physics", **7**, 328 (1958).

[6] Hashimoto, H., and Uyeda, R., *Acta Cryst.*, **10**, 143 (1957).

[7] Frank, F. C., *Acta Cryst.*, **4**, 497 (1951).

[8] Garner, W. E., "Chemistry of the Solid State", 116 (London, 1955).

METALLURGY

Decomposition of the Beta Phase in a Copper–Indium Eutectoid Alloy

SOME optical and electron microscopical observations are reported of certain structures resulting from the decomposition of the beta-phase in a copper–indium alloy of approximately eutectoid composition.

The body-centred cubic beta-phase in the copper–indium system undergoes a eutectoid transformation, the eutectoid composition and temperature being approximately 31·4 wt. per cent indium and 574° C. respectively[1,2]. The product of this transformation is pearlitic, consisting of the alpha- and delta-phases. A detailed metallographic investigation of the isothermal transformation characteristics was made by Spencer and Mack[3]. Transformation at approximately 570° C. was found to occur by the formation of nodules of pearlite, resolvable under the optical microscope. The structure of nodules formed during transformation at 547° C. was not resolved optically, but it was reported that it was not homogeneous. Beta-phase, which had not transformed to pearlite, showed a rough-etching or cross-hatched structure when quenched to room temperature.

In the present work an alloy of approximately eutectoid composition was cast into a graphite mould of 3/8 in. diameter. The ingot produced was homogenized for three days at 650° C., and was then cut into disks of 3/32 in. thickness. Disks were quenched from 650° C. into a salt bath at either 565° C. or 547° C., and after being held for selected times at one of these temperatures, they were quenched into cold water. Disks were also quenched directly from 650° C. into cold water. Metallographic preparation of the heat-treated disks included mounting in 'Bakelite', grinding approximately 0·02 in. from the exposed 3/8-in. diameter surface, polishing with diamond dust and electrolytic etching with 1 per cent chromic acid. Examination was made by both optical and electron microscopy, the latter by means of unshadowed 'Formvar' replicas.

Optical microscopical examination (at × 1,400) of specimens partially transformed at 565° and 547° C. showed nodules which were partly resolvable as pearlite: electron microscopical examination of a number of replicas showed a completely pearlitic structure within the nodules. A portion of a nodule is shown in Fig. 1.

The beta-phase, which had not transformed to pearlite, when quenched, generally showed a rough etching appearance under the optical microscope, although there were variations in structure. Examination of replicas revealed the cellular structure illustrated in Fig. 1: a similar structure was shown by replicas taken from a specimen quenched directly from 650° C. into cold water. It may be concluded that it results from the decomposition

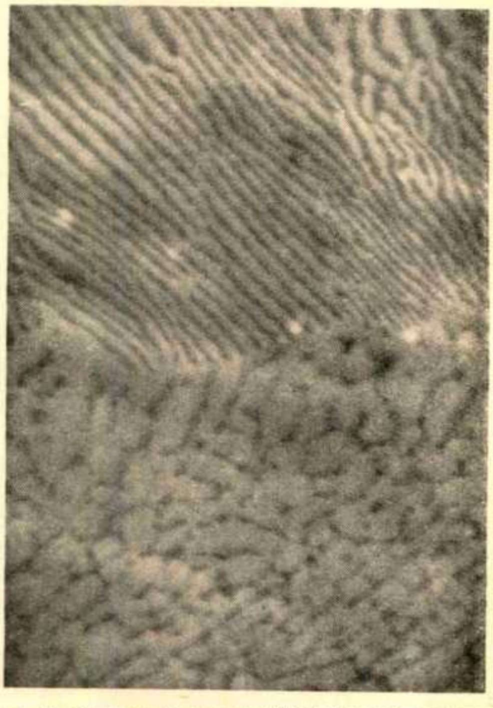

Fig. 1. Replica from specimen partially isothermally transformed at 547° C., showing a portion of a pearlite nodule with the adjacent region of quenched beta-phase (× about 25,000)

of the beta-phase during quenching. The cellular structure bears some resemblance to that observed under the optical microscope in copper–aluminium alloys which have been isothermally treated to produce a mixture of the disordered beta-phase and the ordered beta$_1$-phase (ref. 4 and Thomas, D. L., unpublished work). It is of interest to note that the diffraction pattern of the quenched copper–indium beta-phase has been found to contain lines which indicate a superlattice structure[1]. Thus, it seems possible that the cellular structure may be associated with the occurrence of an ordering reaction on quenching.

Optical microscopy showed that in some regions of the quenched beta-phase, immediately adjacent to the edge of the pearlite nodules, the structure was somewhat columnar in appearance. This effect occurred rather irregularly: where such columnar zones were observed they were commonly approximately 2μ in extent. A similar structural feature was reported by Spencer and Mack[3]. Examination of replicas confirmed that the cellular structure in these zones tended to adopt a columnar form. (This effect is not seen clearly in the particular micrograph shown.) The columnar zones may be associated with the presence of a strain gradient in the beta-phase adjacent to the pearlite nodule interface, the strain being due to volume changes accompanying pearlite formation. During quenching from the transformation temperature, the strain gradient might influence the morphology of the decomposition product of the beta-phase. In the examination of the replicas no interface was detected between the matrix of the cellular structure and one of the phases of the pearlite.

Acknowledgments are made to Mr. H. I. Matthews of the Department of Chemical Engineering, Imperial College, for the use of the Siemens Elmiskop I electron microscope in which the replicas were examined, to Dr. D. L. Thomas for helpful discussions, and to Miss P. Martins for assistance with the photographic work.

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[1] Weibke, F., and Eggers, H., *Z. anorgan. Chem.*, 220, 273 (1934).
[2] Hume-Rothery, W., Raynor, G. V., Reynolds, P. W., and Packer, H. K., *J. Inst. Metals*, 66, 209 (1940).
[3] Spencer, C. W., and Mack, D. J., *J. Inst. Metals*, 82, 81 (1953–54).
[4] Smith, C. S., and Lindlief, W. E., *Trans. A.I.M.M.E.*, 104, 69 (1933).

Embrittlement of Tungsten and of Molybdenum

A recent paper from this laboratory[1] described the embrittlement of tungsten wire if it be heated in atmospheres containing even traces of ammonia. This work has been extended and has revealed a fact, unobserved in the original study, which furnishes further information which may be helpful in understanding the mechanism. In earlier experiments, the procedure was to heat a length of wire, leaving untreated ends which, since they had not been embrittled, could be clamped in the tensile testing machine without fear of fracture there. The disadvantage of this procedure is that the specimen under test had regions which reached lower temperatures than the central portion, so that any minimum in the embrittlement/treating-temperature curve would not be shown. This aspect was studied in the present series of tests. In these, wire was first annealed so that primary recrystallization was complete[2], and then reheated in nitrogen/hydrogen mixture containing 0·2 per cent ammonia. The whole specimens were then used for the test, neglecting those which broke in the clamps.

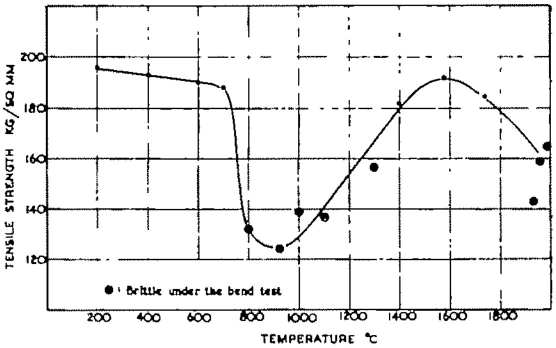

Fig. 1. Effect of heating for 10 min. in 0·2 per cent ammonia on the tensile strength at 20° C. of 180-μ tungsten wire

The results are given graphically in Fig. 1 and show that a minimum embrittlement does indeed occur after heating near 1,600° C.; above this, there is again further embrittlement. Both strength and bend tests were made; the latter were the more subjective but gave clear indications of qualitative changes: specimens which were brittle under this test are also indicated in Fig. 1.

Some recent work which we hope to publish soon has shown that embrittlement of molybdenum wires occurs on heating in presence of elementary nitrogen. While this effect can be important practically, it is quantitatively rather too small to reveal clearly any minimum in the mechanical properties. However, measurements of gas content of treated wires demonstrated that the absorption of nitrogen certainly shows a minimum near 1,300° C., which was not observed in the temperature-range studied by Tury[3], and it may be that there is a parallel mechanism operating for these two similar metals.

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[1] Davis, G. L., *Metallurgia*, 54, 18 (1956).
[2] Davis, G. L., *Metallurgia*, 58, 177 and 228 (1958).
[3] Tury, P., *Math. naturw. Anz. Ungar. Akad. Wiss.*, 57, 571 (1939).

CHEMISTRY

Benedict's Solution as a Developer in Carbohydrate Chromatography

While a large number of reagents have been developed for the differentiation of carbohydrates by chromatography, there is still some difficulty in showing reducing sugars in many biological materials. Many of the developers, such as silver nitrate, tend to become easily reduced and to stain the whole paper, the carbohydrates being shown as dark spots on a similarly stained background. This background stain can be removed[1], but the process is both time-consuming and cumbersome.

For many years Benedict's solution has been used to show the presence of reducing sugars in urine, a positive reaction being obtained in solutions containing less than 0·1 per cent glucose. This solution has been used in this laboratory to develop the spots of reducing sugars in chromatograms run to differentiate galactose and glucose in urines containing sugar. The sugars show up as bright orange spots on a pale blue to white background. The intensity of the spot is directly proportional to the concentration of the carbohydrate, and can be used to give a rough estimate of concentration. Two solvents have been used to prepare the chromatograms. These are butanol/acetic acid/water and water-saturated phenol. This last is used in an atmosphere of ammonia. The solutions are allowed to run in descending chromatography for 24 hr.; 0·01 ml. of standard solutions of 1 per cent glucose and galactose are spotted on the base line. The urines are shaken for 20 min. with 'Amberlite *IR*-120' (H) resin and for a further 20 min. with 'Amberlite *IRA*-400' (OH) resin to remove cations and anions which cause tailing of the spots. 0·02 ml. of urine is spotted on the base line and the chromatogram is prepared as described above. The papers are removed from the developing tanks and dried at 80° C. and dipped into or sprayed with Benedict's solution. I find dipping more effective as it gives a more even distribution of the solution. The chromatograms are then developed by drying at 105° C. for 30 min. The spots are fairly permanent if the papers are kept in the dark after evaluation.

This technique has been used successfully for a wide variety of reducing sugars.

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[1] Benson, A. B., *et al.*, *J. Biol. Chem.*, **196**, 703 (1952).

A New Silicon Boride, SiB_4

THE existence of compounds in the silicon–boron system has been a subject of conjecture for more than fifty years, since the many studies that have been made have produced negative, inconclusive, or contradicting results. Moissan and Stock[1], in the original work on the system, reported that by fusion of the elements they were able to prepare two compounds, SiB_3 and SiB_6. The SiB_3 was obtained as black rhombic plates with a density of 2·52 gm./cm.^{3}, and the SiB_6 as black irregular crystals with a density of 2·47 gm./cm.^{3}. The borides were reported to have high melting points, high hardness, and to be conductors of electricity, properties which prompted future workers to conduct further investigations.

Attempts by Tone[2] and Brewer[3] were unsuccessful. Studies of ternary systems involving silicon and boron[4] were uncertain regarding the silicon–boron binary, although a possible compound was indicated at about 90 per cent boron. Recently, Samsonov and Latysheva[5] have reported the properties of a tetragonal SiB_3 prepared by a method similar to that of Moissan. Their conclusions regarding the identification of the compound are doubtful, however, as Ormont *et al.*[6] have pointed out. Samsonov and Latysheva also prepared a compound SiB_6, which was studied by Zhuravlev[7], who reported a structure isomorphous with that of CaB_6.

Concurrent with the Russian work, Cline[8] prepared single crystals of SiB_6 and measured many of the physical properties[9]. The crystallographic data[10] indicate that the SiB_6 is orthorhombic and the measured density of 2·43 gm./cm.^{3} is much higher than the value of 2·15 gm./cm.^{3} reported by Zhuravlev for the cubic phase. Further experiments in the system involving the fusion of SiB_6 and silicon yielded a new phase which occurred as rhombs and hexagonal plates. The crystals are reddish-brown under reflected light and have a measured density (sink-float) of 2·44 gm./cm.^{3}. Chemical and spectrographic analyses indicate the formula SiB_4.

Rotation and Weissenberg zero- and upper-level photographs of single crystals of various forms of the SiB_4 indicate a rhombohedral lattice with a = 5·52 A. and α = 69·1°. The corresponding hexagonal cell constants are A = 6·35 A. and C = 12·69 A. There are three molecules of SiB_4 in the rhombohedral unit cell. The probable space groups are $R32$, $R\bar{3}m$, and $R3m$. The density calculated from these data is 2·41 gm./cm.^{3}.

The physical properties of SiB_4 are under investigation, and these properties, as well as the details of the preparation, will be reported at a later date. Intensities are being measured, and a detailed examination of the structure will be made. SiB_4 appears to have some structural features in common with B_4C (ref. 11), and with various forms of elemental boron[12,13], and a comparison of the structures should prove extremely interesting.

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[1] Moissan, A., and Stock, A., *C.R. Acad. Sci., Paris*, **131**, 139 (1900).

[2] Tone, F., *Indust. Eng. Chem., Ind. Ed.*, **30**, 252 (1938).

[3] Brewer, L., Sawyer, D. L., Templeton, D. H., and Dauben, C. H., *J. Amer. Ceram. Soc.*, 173 (1952).

[4] Nowotny, H., Dimakopoulou, E., and Kudielka, H., *Monatsh. Chem.*, **88**, No. 2, 180 (1957).

[5] Samsonov, G. V., and Latysheva, V. P., *Doklady Akad. Nauk., S.S.S.R.*, **105**, No. 3, 499 (1955).

[6] Gurevich, M. A., Epel'baum, V. A., and Ormont, B. F., *Zhur. Neorg. Khim.*, **2**, 206 (1957).

[7] Zhuravlev, N. N., *Kristallografia*, **1**, 666 (1956).

[8] Cline, C. F., *Nature*, **181**, 476 (1958).

[9] Cline, C. F., *J. Electrochem. Soc.*, **106**, 322 (1959).

[10] Adamsky, R. F., *Acta Cryst.*, **11**, 744 (1958).

[11] Clark, H. K., and Hoard, J. L., *J. Amer. Chem. Soc.*, **65**, 2115 (1943).

[12] Hoard, J. L., Hughes, R. E., and Sands, D. E., *J. Amer. Chem. Soc.*, **80**, 450 (1958).

[13] McCarty, L. V., Kasper, J. S., Horn, F. H., Decker, B. F., and Newkirk, A. E., *J. Amer. Chem. Soc.*, **80**, 2592 (1958).

Products of Decomposition of Lead Azide

PREVIOUS work on the decomposition of alpha-lead azide have been performed at temperatures close to its ignition temperature[1,2]. The assumption has been made that the products of such decompositions have been only lead metal and nitrogen gas. The present communication describes the results of a study on the solid material obtained from the low-temperature thermal decomposition of alpha-lead azide.

The material used was a finely divided form of lead azide, prepared in as pure a form as possible. Crystal

Table 1. ALPHA-LEAD AZIDE HEATED AT 175° C. FOR 215 HR.

Copper $K\alpha$-radiation : product, $PbN_{3·33}$ (γ-phase) $a_0=12·25_4$A. Cubic indexed.

d obs.	Relative intensity	sin θ (obs.)	$\sin^2\theta$ (obs.)	$\sin^2\theta$ (calc.)	$h^2+k^2+l^2$	h	k	l
8·67	5·4	0·0889	0·0079	0·0079	2	1	1	0
7·08	19·6	0·1089	0·0119	0·0119	3	1	1	1
4·09	10·9	0·1885	0·0355	0·0356	9	300	;	221
3·88	10·9	0·1987	0·0395	0·0396	10	3	1	0
3·06	100	0·2519	0·0635	0·0633	16	4	0	0
2·74	38·0	0·2814	0·0792	0·0792	20	4	2	0
2·67	22·8	0·2887	0·0834	0·0832	21	4	2	1
2·61	19·6	0·2954	0·0874	0·0871	22	3	3	2
2·40	5·4	0·3212	0·1032	0·1030	26	510	;	431
2·36	7·6	0·3267	0·1067	0·1069	27	511	,	333
2·28	5·4	0·3381	0·1143	0·1148	29	520	,	432
1·91	19·6	0·4036	0·1629	0·1624	41	621	;	540, 443
1·87	5·4	0·4122	0·1699	0·1702	43	5	3	3
1·67	10·9	0·4616	0·2131	0·2138	54	721	,	633, 552
1·61	10·9	0·4788	0·2293	0·2297	58	7	3	0
1·59	3·3	0·4848	0·2350	0·2335	50	731	;	553

Table 2. CHEMICAL ANALYSIS OF HEAT-TREATED ALPHA-LEAD AZIDE

Sample No.	Treatment	Nitrogen (per cent)	Empirical formula	Phases	Colour
1	Untreated	28·90	$PbN_{6·0}$	α-lead azide	white
2	Heated 150° C.	21·36	$PbN_{4·25}$	phase 1 plus phase 2	light yellow
3	Heated 175° C	17·80	$PbN_{3·30}$	phase 2 plus phase 3	intense yellow
4	Heated 200° C.	16·86	$PbN_{3·01}$	phase 2 plus phase 3	yellow-brown

Table 3

Compound	Lattice type and symmetry	Cell dimensions (A)	Atoms per unit cell	Space group	Density (gm./cm.³)
Lead azide decomposition product, $PbN_{3·3}$	Unknown deformed primitive, cubic	$a_0=12·25_4$ $c_0<a_0$	$Z=25$		5·633
Metal, Pb	A1 cubic, face-centred	$a_0=4·9396$	$Z=4$	O^5_h, $Fm3m$	11·341
PbO	B10 (tetragonal)	$a_0=3·972$ $c_0=5·023$	$Z=2$	D^7_{4h}, $P4/nmm$	9·35
Pb_3O_4	Unknown, tetragonal	$a_0=8·80$ $c_0=6·56$	$Z=4$	D^{13}_{4h}, $P4/mbc$ or D^7_{2d}, $P\bar{4}b2$	8·95
$PbCO_3$	Unknown, orthorhombic	$a_0=5·166$ $b_0=8·468$ $c_0=6·146$			

modifiers of all kinds were excluded. Cation impurities such as sodium, calcium, magnesium, iron, silicon and silver totalled less than 0·1 per cent. The mean particle size of the crystals was 7μ.

After thermal decomposition of the azide in fused silica crucibles, the solid residue was examined by X-ray crystallography using copper $K\alpha$-radiation. Both Debye–Scherrer and X-ray diffractograms were obtained. Samples weighing 0·250 gm. were decomposed under ambient pressure and temperatures of 150°, 175° and 200° C.

When the decomposition of lead azide is followed thermogravimetrically at a series of constant temperatures, weight-loss versus time curves like those in Fig. 1 are obtained. These results suggest that at least two phases in the decomposition of alpha-lead azide can be obtained, but the results do not exclude the possibility that the pauses in the curve may be the result of some unknown diffusion process. The results of a decomposition at 175° C., permitted to take place for a period of 230 hr., are shown in Fig. 2. The fact that diffusion is important in the process is evident from a comparison of the co-ordinates of the beginning of the first plateau in this figure and the same curve in Fig. 1.

In order to obtain more information on the nature of the pauses in the decomposition curve, decompositions at 175° C. were carried out for 215 hr. The result in Fig. 2 is representative. The constancy in the loss in weight at the first plateau is noteworthy.

The material which resulted from such decompositions had an intense yellow colour. X-ray diffractogram and Debye-Scherrer patterns were obtained on this material (see Table 1). The reflexions observed have been indexed as primitive cubic and they show excellent agreement between the observed and calculated $\sin^2\theta$ values. In addition, the calculation of the cell parameters suggests that the unit cell is not a perfect cube, since the c_0/a_0 ratio is not exactly 1.

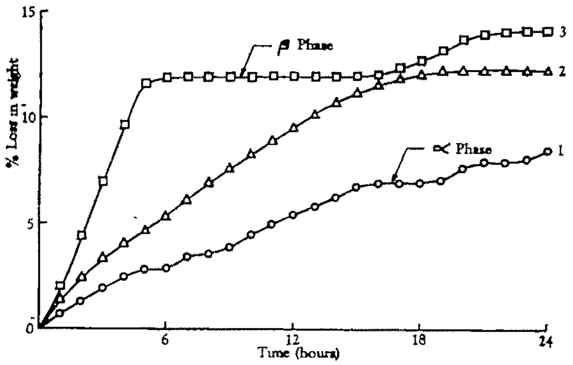

Fig. 1. Thermogravimetric study of decomposition of α-lead azide. (1) Decomposition at 150° C, α-phase ; (2) decomposition at 175° C. ; (3) decomposition at 200°C.

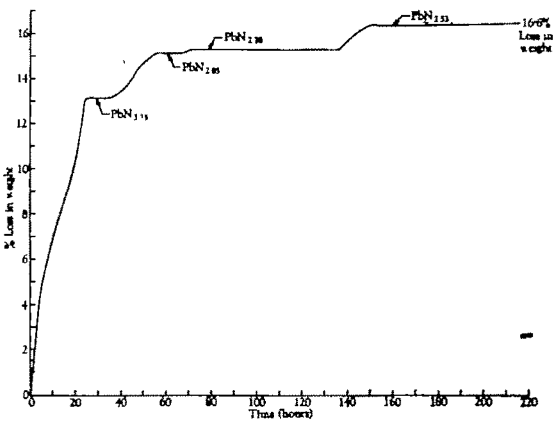

Fig. 2. Decomposition curve of α-lead azide at 175° C.

X-ray analyses of products of intermediate decompositions such as those characterized by the treatments in Fig. 1 suggest that a mixture of phases is present. Relative intensities are presented in Fig. 3 as functions of the reflexion angles. Nitrogen content by chemical analysis of these phases is shown in Table 2. Additional analyses for both lead and nitrogen were performed on a duplicate of sample 4 of this table. The lead was precipitated as lead sulphate and nitrogen determined by Dumas's method. The results in duplicate are :

Lead 81·98 per cent	Nitrogen 16·53 per cent
82 03	16·65
av. 82·00	av. 16·59

The material balance shows that oxygen can be present in the decomposition product only to the extent of 1·4 per cent if at all.

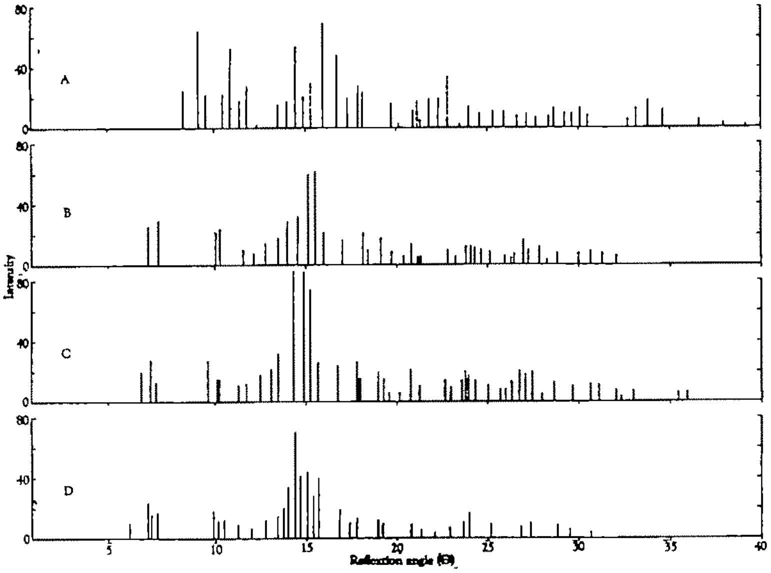

Fig. 3. Comparison of X-ray reflexions of untreated and heat-treated α-lead azide (*A*) Untreated α-$Pb(N_3)_2$; (*B*) $PbN_{4\cdot33}$ α-$Pb(N_3)_2$ heated for 24 hr. at 150° C. ; (*C*) $PbN_{3\cdot33}$ α-$Pb(N_3)_2$ heated for 24 hr. at 175° C. ; (*D*) $PbN_{3\cdot09}$ α-$Pb(N_3)_2$ heated for 24 hr. at 200° C

These results, plus an examination of data for probable materials which might result from heating alpha-lead azide in air as compiled in Table 3, suggests that under these conditions alpha-lead azide decomposes in a step-wise fashion through several distinct phases having varied lead to nitrogen ratios. The clarity of the diffractograms has been rewarding, and the very sharp lines exhibited have given no reason for assuming disorder in the structures. Work to confirm these results in more detail is in progress.

We thank Mr. J. J. Campisi for taking the X-ray diffractograms and Debye–Scherrer patterns : and Mr. C. Campbell for the thermogravimetric work.

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[1] Griffiths, P. J. F., and Groocock, J. M , *J. Chem. Soc.*, 3380 (1957).
[2] Garner, W. E., and Gomm, A. S., *J. Chem. Soc.*, 2123 (1931).
[3] Garner, W. E., *Proc. Roy. Soc.*, A, 246, 203 (1958).

Molecular Structure of Neopentane

WE have recently studied the molecular structure of pivalonitrile[1], $(CH_3)_3CCN$, and have found the central C—C bond to be 1·46 ± 0·02 A. The carbon skeleton in pivalonitrile is similar to that in neopentane and, therefore, a comparison of the C—C distance in neopentane with the central C—C distance in pivalonitrile should be a measure of the effect of an adjacent triple bond on the C—C distance. It was also considered interesting to see if the C—C distance in neopentane would be longer than in ethane due possibly to steric repulsions. The only parameters available for neopentane were from the early visual work of Pauling and Brockway[2] and a re-investigation of the structure of this molecule seemed timely.

The sample used for the diffraction photographs was obtained from Phillips Petroleum Co. It was characterized as Phillips's research grade with a mole per cent purity of 99·92. Electron diffraction photographs were taken, using an r^3 sector, at camera distances of 25·10 cm. and 10·19 cm. employing electrons of wave-length 0·05452 A. and Kodak lantern slide plates. Exposure times of 10 sec. for the long camera distance and 30–45 sec. for the short camera distance were used. Three patterns at the short camera distance and two at the long camera distance were chosen for the visual measurement of ring diameters and estimation of intensities. The visual results from the sectored plates extended from $q = 17$ to $q = 100$.

A radial distribution curve was calculated from the sector-visual intensity data[3]. The data for the range $q = 0$ to $q = 17$ were supplied from a theoretical curve[3].

The radial distribution curve (Fig. 1) shows three peaks and a few shoulders. The peak at 1·09 A. corresponds to the bonded C—H distance and the one at 1·54 A. corresponds to the bonded C—C distance The peak at 2·51 A. corresponds to the non-bonded C···C distance. The shoulder at 2·17 A. is attributed to the short non-bonded C···H distance. The shoulder at about 2·75 A. is due to the intermediate non-bonded C···H distance and the peak at 3·46 A. corresponds to the long non-bonded C···H distance. It is interesting to note that there is a small shoulder at about 1·8 A. which is caused by the short non-bonded H···H distance. The amplitudes of vibration of a few atom pairs were estimated by Karles's method[4,5]. The radial distribution results are summarized in Table 1.

Table 1. RESULTS FROM THE RADIAL DISTRIBUTION CURVE

Distance	Peak position (A.)	Δr_{ij}	Area[c]	$\frac{nZ_iZ_j}{r_{ij}}$
C—H	$1\cdot08_9$	0 075	90	88
C—C	$1\cdot54_4$	0·05	100	100
C·· H (short)	$2\cdot16_4$[a]	—		
C·· C	$2\ 51_4$[b]	0·08	201	206
C·· H (intermediate)	—	—		
C·· H (long)	$3\cdot45_8$	$0\ 20_8$	26	28

[a] This is consistent with a CCC angle of 109° 28′.
[b] Appears as a shoulder at about 2·75 A.
[c] Areas of the peaks were approximated to bell shapes.

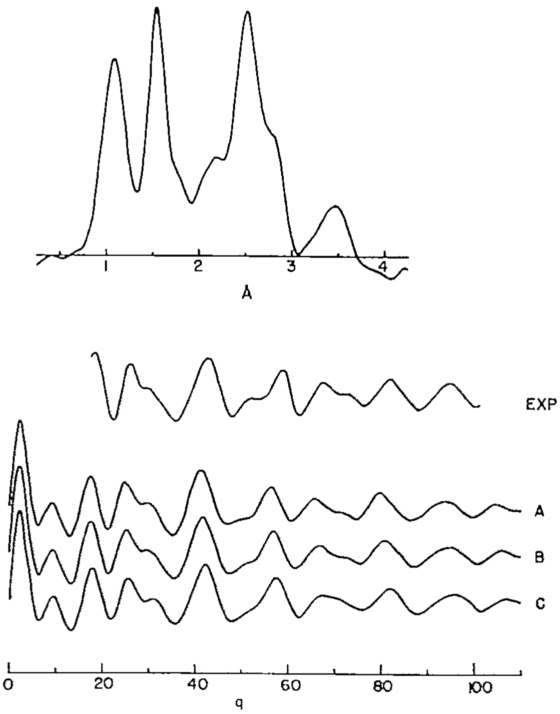

Fig. 1. The radial distribution curve and the experimental and theoretical intensity curves for neopentane

A correlation procedure has also been carried out. Six theoretical models with bonded C—C distances of 1·48 A. (*E*), 1·50 A. (*D*), 1·52 A. (*C*), 1·54 A. (*B*), 1·56 A. (*A*), and 1·58 A. (*AA*) were calculated. The bonded C—H distance and the CCC angle were maintained constant at 1·09 A. and 109·5° respectively. The hydrogen atoms were kept as far as possible from each other. Of these models all are qualitatively acceptable with the possible exception of the 1·48 A. model which is a borderline case. A model calculated with a bonded C—C distance of 1·54 A. assuming free rotation was found completely unacceptable. The quantitative electron diffraction results for the acceptable models are given in Table 2 and the parameters from the acceptable models are summarized in Table 3.

Table 2. QUANTITATIVE ELECTRON DIFFRACTION RESULTS FOR NEOPENTANE

Max.	Min.	q_{obs}	q_{calc}/q_{obs}					
			AA	*A*	*B*	*C*	*D*	*E*
3		17·50	0 985	0 996	1·005	1·018	1·037	1·051
	3	21·77	0·955	0·973	0 986	0 996	1·016	1 028
4		25 16	0·980	0·991	1 008	1 014	1 022	1·032
6		41·80	0 970	0 988	0·995	1 010	1 018	1·034
	6	47·24	0·984	0·999	1·007	1·022	1·030	1·044
8		57·99	0·957	0 969	0·979	0·991	1·002	1·011
	8	61·61	0 971	0·984	1 000	1 015	1 032	1·043
9		66·66	0 969	0 983	1 001	1·017	1 033	—
11		81·14	0·969	0·980	0·993	1·005	1·018	1·034
	11	87·08	0·981	0·991	1·001	1·011	1·023	1·035
12		93·83	0 987	0·997	1·009	1 021	1·033	1 044
	12	99·62	0 988	0·999	1·012	1·023	1·035	1·050
Mean q_{calc}/q_{obs}			0·975	0·987	1·000	1·012	1·025	1·037
Average deviation from the mean			0·010	0·008	0·008	0·008	0·008	0·009

Table 3. PARAMETERS FROM ACCEPTABLE MODELS

	AA	*A*	*B*	*C*	*D*	*E*
C—H	1 06$_3$	1·07$_8$	1 09$_0$	1 10$_8$	1 11$_7$	1·13$_8$
C—C	1 54$_1$	1·54$_8$	1·54$_8$	1·53$_8$	1 53$_8$	1·53$_3$

The final parameters of the molecule from this study are:

$$\text{C—C} = 1{\cdot}54 \pm 0{\cdot}01 \text{ A.}$$
$$\angle\text{CCC} = 109{\cdot}5 \pm 1°$$

No attempt was made to determine the C—H distance as any such determination would result in an uncertainty of at least ±0·05 A.

It is interesting to note that the C—C distance in neopentane is equal to the value of 1·54 A., which has usually been accepted as the normal C—C single-bond distance. Within the estimated uncertainty, this distance is also consistent with the value of 1·533 A. proposed by Bartell[6] as the normal C—C single bond distance in straight-chain hydrocarbons. The interatomic distances obtained in this investigation are in agreement with those obtained earlier by Pauling and Brockway[2], but the present work is regarded as more accurate.

We wish to thank the Phillips Petroleum Co. for providing the sample of neopentane. This communication contains material from the master's thesis of one of us (C. L.).

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[1] Livingston, R. L., and Rao, C. N. R., *J. Amer. Chem. Soc.*, 81, 3584 (1959).

[2] Pauling, L., and Brockway, L. O., *J Amer. Chem. Soc.*, 59, 1223 (1937).

[3] Livingston, R. L., Rao, C. N. R., Kaplan, L. H., and Rocks, L., *J. Amer. Chem. Soc.*, 80, 5368 (1958).

[4] Karle, J., and Karle, I. L., *J. Chem. Phys.*, 17, 1052 (1949).

[5] Karle, I. L., and Karle, J., *J. Chem. Phys.*, 18, 957 (1950).

[6] Bartell, L. S., *J. Amer. Chem. Soc.*, 81, 3497 (1959).

Crystallization of Non-Racemic Mixtures of the Isomers of Serine

IF the enantiomorph (unlabelled) of a labelled isomer is available, the maximum contamination of the particular labelled compound by its enantiomorph may be assessed by a crystallization method. Repeated crystallization of a mixture of enantiomorphs in which one (unlabelled) is present in large excess of the other (labelled) would be expected to lead to a purification of the more abundant isomer; hence, the presence of isotope in the purified abundant isomer should represent contamination of the labelled antipode. Thus, when enzymatically prepared[1,2] D-glutamic acid-2-^{14}C was diluted about 2,000-fold with carrier L-glutamic acid and the mixture crystallized, via the hydrochloride and the free amino-acid, to constant specific activity, it could be calculated that the maximum amount of contaminant L-glutamic acid-2-^{14}C was less than 1 per cent[3].

We have resolved DL-serine-3-^{14}C (obtained from Nuclear-Chicago Corp.) by the selective enzymatic hydrolysis of the N-chloroacetyl derivative[3]. To estimate the purity of the labelled D- and L-serine samples, the latter were diluted about 2,000-fold with carrier D- or L-serine and subjected to repeated recrystallization from water or ethanol–water mixtures. Aliquots of the various crystalline products obtained were assayed for carbon-14[4]. The results are presented in Table 1. Surprisingly, when the labelled serine isomers were diluted with their respective antipodes (^{14}L + D, or ^{14}D + L), successive

Table 1. SPECIFIC ACTIVITIES AFTER SUCCESSIVE RECRYSTALLIZATIONS OF D- OR L-SERINE-3-^{14}C DILUTED ABOUT 2,000-FOLD WITH CARRIER D- OR L-SERINE*

No. of recrystallization	Range of yield	μc./m.mole†			
		^{14}L+D	^{14}L+L	^{14}D+L	^{14}D+D
A Serine (first series)					
4	40–52‡	913	492	898	494
5	76–80§	1,122	485	1,139	494
6	64–76§	1,545	462	1,650	478
7	20–24§	5,200	280	5,740	331
B Serine (second series)					
Dried aliquot	—	470	487	493	483
1	93–96‡	495	485	500	476
2	12–18§	1,710	368	3,390	387
3	30–45§	—	—	5,900	—
C *p*-Toluenesulphonyl derivative of product of first crystallization in *B* (above)					
2		22	503	22	467
3		3·8	—	8·6	—
4		—	—	5·9	—

* Obtained commercially: $[\alpha]^{27}_{D}$, − 11·8 ± 1° (D-serine).

$[\alpha]^{27}_{D}$, + 12·8 ± 1° (L-serine)

† Corrected for dilution with carrier. Therefore, the results give the relative specific activities of each precipitate.

‡ Per cent (by weight) of original mixture recovered.

§ Per cent (by weight) of previous crystallization recovered.

recrystallization led to an increase in specific activity, the increase being approximately inversely proportional to the recovery (by weight) during a given crystallization. Thus, in the case of ^{14}D + L (Series *A*, seventh recrystallization), 4·2 per cent of the original serine contained 49 per cent of the original carbon-14, an enrichment of isotope of more than eleven-fold. Repeated recrystallization of ^{14}L + L or of ^{14}D + D gave products of fairly constant specific activity except when the recovery of serine was low, in which case the specific activities fell markedly.

These unusual results can be explained only by assuming that the carrier D- and L-serine used were contaminated with several per cent of their respective enantiomorphs, and that during recrystallization the contaminants were preferentially precipitated. Apparently, the DL-component of each mixture was almost quantitatively precipitated under conditions in which the L- or D-isomer was quite soluble. The lesser solubility of DL-serine as compared with D- or L-serine[5] is in accord with this hypothesis, although the difference in solubilities does not afford a complete explanation of the results.

Optical rotations, determined spectrophotometrically, support the above hypothesis. (Optical rotations were determined by using a Keston polarimeter attachment to a Beckman Model *DU* spectrophotometer (Standard Polarimeter Co., Hackensack, New Jersey, U.S.A.). All rotation measurements were made at 589 mμ using 5 *N* hydrochloric acid as solvent (temperature 24–27° C.)). The specific rotations of the carrier D- and L-serine were, respectively, − 11·8 ± 1° and + 12·8 ± 1°. Specific rotations ranging from 0 to 4·60 ± 3° were obtained on the final precipitates of the two serine recrystallization series (Table 1). Greenstein *et al.*[2] have reported specific rotations for pure serine isomers (in 5 *N* hydrochloric acid) of about 15°.

In contrast to the results with serine, recrystallization of the *p*-toluenesulphonyl derivatives gave a purification of the more abundant isomer. Thus, in the case of ^{14}D + L (series *B* and *C*), recrystallization as serine yielded a precipitate the specific activity of which was 1,000 times that obtained after recrystallizing the *p*-tosyl derivative of the same serine sample. Results obtained with the *p*-tosyl derivatives prove that the D- and L-serine-3-^{14}C samples contained no more than about 1 per cent of their respective enantiomorphs.

The technique described above for assessing a slight degree of racemization in labelled D- or L-compounds appears to be a valid and valuable one. However, in instances where the racemate is less soluble than the D- or L-isomers, large errors are possible if the isomers used for dilution are not pure. In this connexion we found that a single crystallization of D-serine-3-^{14}C, diluted 5,000-fold with L-serine of higher optical purity ($[\alpha]^{27}_{D}$ = + 14·6 ± 1°) than that originally used, yielded results similar to those obtained with the *p*-tosyl derivatives.

The results also illustrate that recrystallization of an optical isomer contaminated with only a few per cent of its enantiomorph may lead to a concentration rather than removal of the contaminant.

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[1] Meister, A., Sober, H. A., and Tice, S. V., *J. Biol. Chem.*, 189, 577 (1951).

[2] Birnbaum, S. M., Levintow, L., Kingsley, R. B., and Greenstein, J. P., *J. Biol. Chem.*, 194, 455 (1952).

[3] Wilson, W. E., Hill, R. J., and Koeppe, R. E. (unpublished results).

[4] Koeppe, R. E., and Hill, R. J., *J. Biol. Chem.*, 216, 813 (1955).

[5] Lange, N. A., "Handbook of Chemistry", ninth ed., 676 (Handbook Pub., Inc., Sandusky, Ohio, 1956).

A New Route to 1-Substituted Pyrazole-4-Carboxylic Acids

1-PHENYLPYRAZOLE-4-CARBOXYLIC acid has been hitherto most successfully synthesized by chloromethylation of 1-phenylpyrazole and subsequent oxidation of the 4-chloromethyl-1-phenylpyrazole[1], or by hydrolysis of the 4-cyano derivative formed from the anti-oxime of 4-formyl-1-phenylpyrazole[2]. A synthesis of this acid has now been achieved by the hydrolysis of 4-cyano-1-phenylpyrazole formed by a Sandmeyer type reaction from 1-phenylpyrazole-4-diazonium chloride. Replacement of the pyrazole-4-diazonium group by nitrile has hitherto been unsuccessful[3,6].

The starting material, 4-nitro-1-phenylpyrazole, was prepared in quantitative yield by the method of Hill and Torrey[4]. Reduction by hydrazine catalysed by Raney nickel[5] yielded 4-amino-1-phenylpyrazole (97 per cent). This reagent we found to be an excellent reductant for 4-nitropyrazoles. The following pyrazoles: 4-nitro-1-2′-quinolyl-, 1-2′-benzthiazolyl-4-nitro-, and 4-nitro-1-2-5-trimethyl- have also been reduced to the corresponding amines in yields varying between 87 and 100 per cent (unpublished work).

4-Amino-1-phenylpyrazole was diazotized and added to a potassium cuprocyanide solution at 80°.

Ether extraction gave a 56 per cent yield of 4-cyano-1-phenylpyrazole, m.p. 94–95° (Lit.[3] 95°). Found: C, 70·8; H, 4·4; N, 24·8. Calc. for $C_{10}H_7N_3$: C, 71·1; H, 4·1; N, 24·8 per cent. A quantitative yield of 1-phenylpyrazole-4-carboxylic acid, m.p. 221–222°, was obtained on hydrochloric acid hydrolysis of the 4-cyanopyrazole.

The overall yield of 1-phenylpyrazole-4-carboxylic acid by this synthesis is greater than 50 per cent and the route should provide easy access to other 1-substituted pyrazole-4-carboxylic acids.

There is but one reference in the literature to a similar substitution by this method of a pyrazole-4-diazonium group by halogen and the yields were poor[6]. We have found that, utilizing cupric salts as catalysts at elevated temperatures, halogen can be introduced in good yield, for example, 1-phenylpyrazole-4-diazonium bromide gives 4-bromo-1-phenylpyrazole in 86 per cent yield.

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[1] Finar, I. L., and Godfrey, K. E., *J. Chem. Soc.*, 2293 (1954).
[2] Finar, I. L., and Lord, G. H., *J. Chem. Soc.*, 3314 (1957).
[3] Knorr, L., *Ber.*, 28, 714 (1895).
[4] Hill, H. B., and Torrey, J., *Amer. Chem. J.*, 22, 89 (1899).
[5] Balcom, D., and Furst, A., *J. Amer. Chem. Soc.*, 75, 4334 (1953).
[6] Morgan, G. T., and Ackerman, I., *J. Chem. Soc.*, 123, 1308 (1923).

BIOCHEMISTRY

3 : 3′-Diamidinocarbanilide: a New Drug Active against Babesial Infections

Quinuronium sulphate[1] is the commonly employed treatment for British bovine redwater and a single injection of 1 mgm. of this drug per kgm. of body-weight is effective in a high proportion of cases. But quinuronium sulphate has the disadvantage that even in therapeutic doses it produces toxic effects associated with parasympathetic stimulation. The diamidines, phenamidine and pentamidine, as their di-isethionates, and Berenil are well tolerated and are also used for the treatment of bovine babesiasis, but they are less active than quinuronium sulphate.

During the search for a new drug which would be at least as effective as quinuronium sulphate but without its toxic effects, a large number of compounds has been screened for activity against *Babesia rodhaini* in mice, and the most promising of these were tested against *Babesia divergens* in splenectomized calves. One of the best of these was M and B 5062*A* — 3 : 3′ - diamidinocarbanilide di-isethionate, which was active against *B. divergens* in calves and produced none of the toxic effects associated with quinuronium sulphate. This activity of the 3 : 3′-diamidine was unexpected because of the inactivity of the corresponding 4 : 4′-diamidine. 3 : 3′-Diamidinocarbanilide di-isethionate[2] was prepared, among other methods, by the usual Pinner reaction on the corresponding dinitrile. It is a white, crystalline, anhydrous solid, m.p. 209° C. (decomposing at 256° C.) (found: N, 15·1; S, 11·6 per cent. $C_{15}H_{16}ON_6,2C_2H_6O_4S$ requires: N, 15·3; S, 11·7 per cent), and is approximately 100 per cent w/v soluble in water.

The acute toxicity (*LD*50) of M. and B. 5062*A* in mice was found to be 120 mgm./kgm. subcutaneously compared with 8 mgm./kgm. for quinuronium sulphate, while a comparison of the two compounds in non-infected, splenectomized calves after subcutaneous injection at a site behind the shoulder showed that the maximum tolerated dose of M. and B. 5062*A* was greater than 40 mgm./kgm., and that of quinuronium sulphate less than 4 mgm./kgm. There were no obvious symptoms of toxicity in the calves that received this dose of M. and B. 5062*A*. During the therapeutic activity tests two calves developed mild ataxia which lasted for about 3 hr. after treatment with a dose of 40 mgm./kgm.; otherwise no symptoms of systemic toxicity were observed.

In the therapeutic activity tests, seventeen calves infected, after splenectomy, with *B. divergens* were treated subcutaneously with M. and B. 5062*A* at doses between 3 and 40 mgm./kgm. body-weight. With all doses the intensity of the parasitæmia was reduced within 24 hr. and complete clearance was obtained within three days. In all the calves treated with 5 mgm./kgm. or higher doses, the hæmoglobinuria was cleared in 24 hr.

Further details, including a report on field investigations, will be published in due course.

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[1] Kikuth, W., *Zbl. Bakt.*, 135, 135 (1935).
[2] May and Baker, Ltd., and Berg, S. S., Appl. No. 40428/58.

Metabolism of 1-^{14}C Lignoceric Acid in the Rat

Lignoceric acid, the straight-chain, saturated C_{24} fatty acid, occurs in sphingolipids in amide linkage with the amino-alcohol sphingosine[1,2]. It has not been found in the ester linkages of neutral fat or glycerophosphatides, and very little information is available on its metabolism. In the work reported here, lignoceric acid labelled with ^{14}C carboxyl was administered to rats and its metabolic fate studied.

Lignoceric acid was isolated from peanut oil and converted to 1-^{14}C lignoceric acid, via 1-^{14}C tricosyl cyanide (modification of Anker's method for synthesis of myristic acid[3]). Contrary to the fatty acids of 18 carbon atoms or less, lignoceric acid did not form a soluble complex with serum albumin. Paper electrophoresis of a mixture of 1-^{14}C lignoceric acid and serum albumin (0·5–1 equivalent of the acid per equivalent of albumin) showed that the radioactive fatty acid did not migrate with the albumin but remained at the point of origin. 1-^{14}C lignoceric acid was therefore injected as a neutralized aqueous suspension into the tail vein of rats; the various organs were isolated and extracted with a 2:1 chloroform methanol mixture. Respiratory carbon dioxide labelled with carbon-14 was collected in some experiments, and, in others, the livers were removed, dried and extracted successively with acetone, petroleum ether and ethanol. The acetone and petroleum

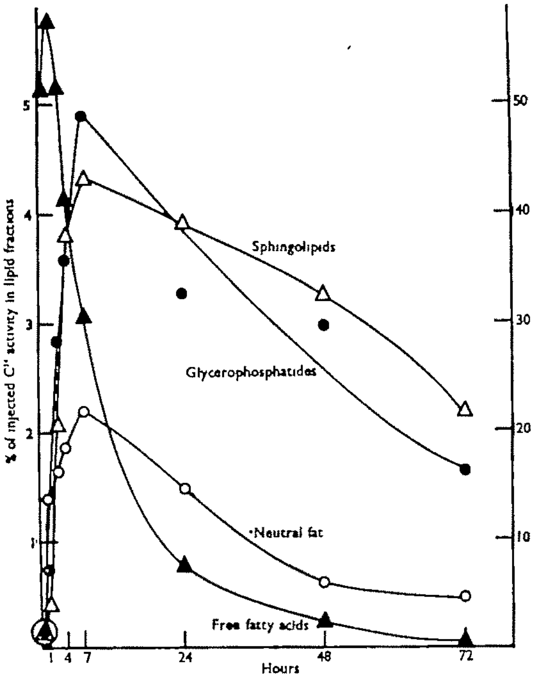

Fig. 1. Incorporation of 1-^{14}C lignoceric acid into rat liver lipids. 6 µmoles of 1-^{14}C lignoceric acid (94,000 c.p.m.) were given by intravenous injection to rats. At the specified times, the animals were, killed and the liver lipids fractionated as described in the text

ether extracts were combined and the lipids separated into a petroleum ether and acetone soluble fraction (that is, neutral fat and free fatty acids) and a petroleum ether soluble, acetone insoluble fraction (that is, glycerophosphatides)[4]. Sphingolipids were isolated from the ethanol extract. The neutral fat was separated from the free fatty acids by passing through a magnesia-'Celite' column[4]. The glycerophosphatides were purified by repeated precipitations of their petroleum ether solutions with acetone and the sphingolipids by repeated precipitations of their ethanol solutions with acetone.

Within 15 min. after injection, about 99 per cent of the 1-^{14}C lignoceric acid was removed from the blood. More than 50 per cent of the injected radioactivity was found in the liver. Small amounts were present in the lungs and spleen ; no radioactivity was found in the brain and only traces in the adipose tissue. A similar pattern of distribution was observed after 24 hr.

About 20 per cent of the radioactivity of the administered 1-^{14}C lignoceric acid appeared in the respiratory labelled carbon dioxide in 7 hr. After an initial lag of about 30 min., the rate of production of labelled carbon dioxide increased, reached a maximum 2 hr. after injection and then declined slowly.

Contrary to the findings with the lower fatty acids[5,6], feeding a stock diet, a diet rich in carbohydrates or fasting, did not influence the formation of labelled carbon dioxide from 1-^{14}C lignoceric acid.

Both the rate of incorporation of 1-^{14}C lignoceric acid into liver lipids and the turnover-rate of these lipids (Fig. 1) were much lower than with palmitic acid[7]. Also, contrary to palmitic acid[7], a large fraction (about 50 per cent) of the injected lignoceric acid was found as the free acid in the liver ; even after 24 hr. about 8 per cent of the free fatty acid was still present in the liver.

A considerable part of the lignoceric acid incorporated into liver lipids was found in the sphingolipids (Fig. 1). However, 1-^{14}C lignoceric acid was also found in the neutral fat and glycerophosphatides. This esterification of lignoceric acid, which has not been observed previously, might be due to the presence, in unphysiological amounts, of free lignoceric acid in the liver.

The results presented indicate that while 1-^{14}C lignoceric acid was metabolized by the rat, its metabolic behaviour differed from that of lower fatty acids, such as palmitic acid. Experiments with tissue preparations are in progress to elucidate further the above findings.

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[1] Deuel, H. J , jun., "The Lipids", 1 (Interscience Publishers Inc., New York, 1951).

[2] Celmer, W. D , and Carter, H. E., *Physiol. Rev.*, 32, 167 (1952).

[3] Anker, H. S., *J. Biol. Chem.*, 194, 177 (1952).

[4] Borgstroem, B., *Acta Physiol. Scand.*, 25, 101 (1952).

[5] Lossow, W. J., and Chaikoff, I. L., *Archives Biochem. and Biophys.*, 57, 23 (1955).

[6] McCalla, C., Gates, H. S., jun., and Gordon, R. S., jun , *Archives Biochem. and Biophys* , 71, 346 (1957).

[7] Stein, Y., and Shapiro. B., *Amer. J. Physiol.*, 196, 1238 (1959).

Ribonucleases in Takadiastase

THE existence of a thermostable ribonuclease in commercial takadiastase was first pointed by Kuninaka[1]. We found afterwards by zone-electrophoresis the existence of at least three enzymes attacking ribonucleic acid in takadiastase: one of them is more active at *p*H 7·5 and the others are more active at *p*H 4·5 [2].

Two of the enzymes, which we named ribonuclease T_1 and ribonuclease T_2, were partially purified by the following procedure: heat treatment, ammonium sulphate fractionation, adsorption to and elution from calcium phosphate gel. Ribonuclease T_1 was further purified by eliminating impurities with active charcoal, and T_2 by chromatography with *DEAE*-cellulose[2,3]. The preparations thus obtained of ribonuclease T_1 and T_2 were shown to be practically homogeneous in zone-electrophoresis.

Ribonuclease T_1 split the internucleotide bonds between 3′-guanylic acid groups and 5′-hydroxyl groups of the adjacent nucleotides with the intermediary formation of guanosine-2′,3′-cyclic phosphate[3–5].

This mechanism was deduced from the following experimental evidence: (1) ribonuclease T_1 digesting yeast ribonucleic acid produces exclusively 3′-guanylic acid (*Gp*) as mononucleotide; (2) the terminal residue of oligonucleotides produced was shown to be exclusively guanylic acid; (3) as an intermediate of the reaction, guanosine-2′,3′-cyclic phosphate ($G_{\text{cyclic P}}$) was obtained with good yield, and it was further hydrolysed to 3′-guanylic acid :

$$
\begin{gathered}
—XpGpXp—XpGpGpXp— \\
\downarrow \text{RNase } T_1 \\
—XpG_{\text{cyclic P}} + Xp—XpG_{\text{cyclic P}} + G_{\text{cyclic P}} + Xp— \\
\downarrow \text{RNase } T_1 \\
—XpGp + Xp—XpGp + Gp + Xp—
\end{gathered}
$$

G, guanosine
X, other nucleosides

Similar experiments were undertaken with ribonuclease T_2, and in this case it was proved that it splits the internucleotide bonds between 3′-adenylic acid groups and 5′-hydroxyl groups of the adjacent nucleotides. However, contrary to the reaction catalysed by ribonuclease T_1, cyclic mononucleotides were not readily obtained[3].

The specificity of ribonuclease T_1 and T_2 is indeed just complementary to that of the well-known pancreatic ribonuclease (ribonuclease 1), at least the main component, component A, which catalyses the cleavage of internucleotide bonds between the 3′-pyrimidine nucleotide groups and the 5′-hydroxyl groups of adjacent nucleotides[6].

Some properties of ribonuclease T_1 and T_2 together with ribonuclease 1 are summarized in Table 1.

Table 1. PROPERTIES OF RIBONUCLEASE T_1, T_2 AND I

	Optimum *p*H	Specificity	Activation and inhibition				References
			Mg++ Ca++	Cu++	EDTA	ICH₂COOH	
T_1	7·5	*Gp*	inhibited	inhibited	activated	activated	(2) (4) (5)
T_2	4·5	*Ap*	inhibited	strongly inhibited	activated	activated	(2) (3)
I	7·6	*Up* *Cp*	activated	inhibited		inhibited	(6)

Ribonuclease T_1 and T_2 catalyse the synthesis of oligonucleotides from guanosine-2′,3′-cyclic phosphate ($G_{\text{cyclic P}}$)[7,8] and adenosine-2′,3′-cyclic phosphate ($A_{\text{cyclic P}}$)[8] respectively. For example, ribonuclease T_1 catalyses the synthesis of guanylyluridine, guanylylcytidine, guanylyladenosine and guanylylguanosine from cyclic guanylic acid and uridine, cytidine, adenosine and guanosine respectively. It catalyses also the polymerization of cyclic guanylic acids. Part of the products are poly- or relatively large oligo-guanylic acids, because they are non-dialysable and give the colour reaction by gallocyanine[9].

$$G_{\text{cyclic P}} + U = GpU \qquad G_{\text{cyclic P}} + C = GpC$$
$$G_{\text{cyclic P}} + A = GpA \qquad G_{\text{cyclic P}} + G = GpG$$
$$nG_{\text{cyclic P}} = GpGpGp\text{—}Gp.$$

Similar reactions were also observed with ribonuclease T_2. These synthetic reactions are analogous to those of oligopyrimidine nucleotides catalysed by ribonuclease I[10]. The generality of such synthetic reactions by certain ribonucleases suggests the possibility of the participation of these enzymes in the intracellular synthesis or reformation of specific ribonucleic acids.

Preparation of oligo- and poly-guanylic acids and cyclic nucleotides by ribonuclease T_1. As mentioned above, ribonuclease T_1 catalyses the polymerization of cyclic guanylic acid. By fractionation on *ECTEOLA*-cellulose column, oligo- and poly-guanylic acids were obtained[8].

K. Sato-Asano and Y. Fujii showed that deaminated yeast ribonucleic acid could be hydrolysed by ribonuclease T_1[11]. By controlled digestion of yeast ribonucleic acid and deaminated yeast ribonucleic acid, guanosine-2′,3′-cyclic phosphate and two new compounds, inosine-2′,3′-cyclic phosphate and xanthosine-2′,3′-cyclic phosphate, were obtained respectively with good yields[12].

Abolition of the streptolysin-forming activity of ribonucleic acid. An interesting biological effect of ribonucleic acid was disclosed by Okamoto's finding that yeast ribonucleic acid stimulated the synthesis of streptolysin ('Streptolysin S') by hæmolytic streptococci[13]. The streptolysin-forming activity of ribonucleic acid was rapidly decreased by ribonuclease T_1[14]. This finding is consistent with the observations which suggest that the activity depends on the consecutive sequence of guanylic acids[15].

Possibility of the determination of nucleotide distribution in ribonucleic acid. As various proteinases have contributed to the determination of amino-acids sequence in protein, ribonucleases with different specificity may be useful for the elucidation of the structure of ribonucleic acid.

In Table 2 the hydrolysis of a hypothetical polynucleotide by ribonuclease I, T_1 and T_2 is presented. By combining the information about the nucleotide sequences of hydrolysis products, the complete nucleotide sequence may be deduced[8].

Table 2. HYDROLYSIS OF A HYPOTHETICAL POLYNUCLEOTIDE

	G–A–U–C–C–G–A–A–G–G–C–U–U–G–A–C–U
Ribonuclease I	*G–A–U C C G–A–A–G–G–C U U G–A–C U*
Ribonuclease T_1	*G A–U–C–C–G A–A–G G C–U–U–G A–C–U*
Ribonuclease T_2	*G–A U–C–C–G–A A G–G–C–U–U–G–A C–U*

Native ribonucleic acids so far obtained are too large and may be a mixture of polynucleotides with different nucleotide sequences. However, by analysing mono- and oligo-nucleotides produced by the three ribonucleases and applying simple probability calculation, we may be able to determine how the nucleotide sequence deviates from that of random distribution. The information thus obtained may be an indication for the specificity of nucleotide distribution of the ribonucleic acid.

In conclusion, we believe that ribonuclease T_1 and T_2 will prove useful for studies of nucleic acids and related problems.

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[1] Kuninaka, A., *J. Agr. Chem. Soc. Japan* **28**, 282 (1954), 29, 52, 797 (1955)

[2] Sato, K., and Egami, F, *J. Biochem.* (*Japan*), **44**, 753 (1957).

[3] Naoi-Tada, M., Sato-Asano, K., and Egami, F., *J. Biochem.* (*Japan*), **46**, 757 (1959).

[4] Sato, K., and Egami, F., *Compt. Rend. Soc. Biol.*, **151**, 1792 (1957).

[5] Sato-Asano, K., *J. Biochem.* (*Japan*), **46**, 31 (1959).

[6] Schmidt, G, "The Nucleic Acids", edited by E. Chargaff and J. N. Davidson, 555 (Academic Press, Inc., 1955).

[7] Sato-Asano, K, and Egami, F., *Biochim. Biophys. Acta*, **29**, 655 (1958).

[8] Egami, F., Naoi, M., and Asano, K., Presented at the Fourth International Congress of Biochemistry, Vienna, 1958.

[9] Scholtissek, C., *Z physiol Chem.*, **309**, 129 (1957).

[10] Heppel, L. A., Whitfeld, P. R., and Markham, R., *Biochem. J.*, **60**, 9 (1955).

[11] Sato-Asano, K., and Fujii, Y., to be published in *J. Biochem.* (*Japan*).

[12] Sato-Asano, K., Fujii, Y., and Egami, F., *Bull. Chem. Soc. Japan*, **32**, 1068 (1959).

[13] Okamoto, H., *Japan J. Med. Sci.*, IV, Pharmacol., **12**, 169 (1939).

[14] Tanaka K, Hayashi, T., and Maekawa, S., *J. Biochem.* (*Japan*), 45, 97 (1958)

[15] Tanaka K., Egami, F., Hayashi, T., and Maekawa, S., *J. Biochem.* (*Japan*), **45**, 593 (1958).

Occurrence of Riboflavin in Sturgeon Skin

LOOMEIFER[1] has reported the presence of a yellow fluorescent pigment in elastin and has commented upon the views of Szent-Györgyi[2] on the function of riboflavin-5-phosphate in metabolism as an energy transmitter. Blair and Graham[3] found riboflavin to be present in the skin of green snakes. These reports of fluorescent materials associated with connective tissues, which are metabolically relatively inert, have prompted this note of the occurrence of riboflavin in sturgeon skin. These observations form part of a survey on non-teleostean fish collagen.

The skin of a young (7 kgm.) sturgeon (*Acipenser sturio*) was mechanically freed from muscle and scales and heated in water, adjusted with acetic acid to *p*H 4·5, for 3 hr. at 60° C. in the dark. Approximately 80 per cent of the collagen was converted into gelatin, which, as a 5·6 per cent (w/v) solution, gave the physical properties shown in Table 1, and compared with gelatins from isinglass (*Acipenser huso huso*), shark skin (*Selachus maximus*) and cod swim-bladder (*Gadus morrhua*) which were prepared under similar conditions.

Table 1. PROPERTIES OF FISH GELATINS

	gelatin 5·6% (w/v)	
	rigidity dynes/cm.[3] at 10° C.	viscosity cp. at 40° C.
Sturgeon skin	25,000	5·2
Isinglass	60,000	5 8
Shark skin	45,000	4·3
Cod swim-bladder	*c.* 100	4·0

The gelatin solution from the sturgeon skin was unusual in that it was associated with a yellow pigment showing a strong green fluorescence. The fluorescence was destroyed by sunlight. The absorption spectrum of this gelatin, plotted between 380 and 580 mμ, was found to be similar (Fig. 1) to that for an acid-processed pig skin gelatin to which riboflavin had been added. In addition, when the sturgeon skin gelatin was irradiated with an ultraviolet source for 30 min. and the absorption curve plotted, it was found that the hump shown by curve *D*, Fig. 1, had been removed so as to resemble curve *B*. The plot of the differences in absorption between the irradiated and non-irradiated forms is shown in Fig. 2. The maximum at approximately 455 mμ corresponds to the maximum given by riboflavin. It seems likely, therefore, that riboflavin is the colour component of this sturgeon gelatin.

It remains to be seen whether the presence of riboflavin in sturgeon skin is a pigmentation phenomenon or whether it is a metabolic factor involved in the laying down of collagen in a young animal.

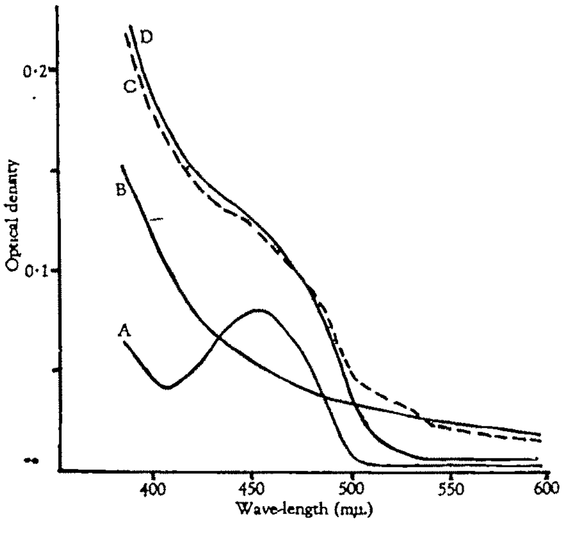

Fig. 1. Absorption spectra: curve *A*, riboflavin; curve *B*, pig skin gelatin; curve *C*, pig skin gelatin with added riboflavin; curve *D*, sturgeon skin gelatin

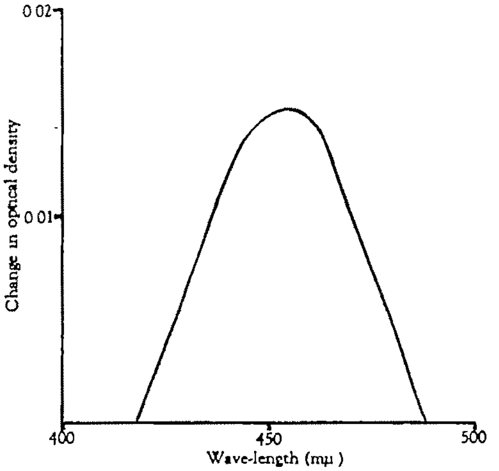

Fig. 2. Changes in the optical densities of sturgeon skin gelatin occurring after ultra-violet irradiation

I am most grateful to H.M. The Queen and to the Comptroller of Supplies, Buckingham Palace, for arranging the gift of a sturgeon.

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[1] Loomeifer, F. J., *Nature*, 182, 182 (1958).

[2] Szent-Gyorgyi, A., "Bioenergetics" (Academic Press, New York, 1957).

[3] Blair, J. A., and Graham, J., *Biochem. J.*, 56, 286 (1954).

Properties and Solubility of Phenolase in Isolated Chloroplasts

THE phenolase complex in green plant tissue has often been assumed to be present only in the chloroplasts[1]; but it has recently been shown that in sugar beet leaf phenolase is also present in the mitochondria and 'soluble' fraction of homogenates. The enzyme associated with chloroplasts was shown to be characterized by a high cresolase activity, the ratio of catecholase to cresolase activities being close to unity. Disruption of such chloroplasts with distilled water increases both activities by 35 per cent[2].

In order to examine further the chloroplast phenolase of sugar beet leaf, we have attempted to extract it by a number of methods. The preparation of chloroplasts and manometric determination of enzymic activities have been described previously[3]. Chloroplasts were macerated by hand in an all-glass homogenizer with the solutions described in Table 1, left to stand for the time indicated and then separated by centrifugation for 5 min. at 3,700*g*. All the preparations were carried out in a cold room at − 1° C.; the acetone had previously been cooled to − 20° C.

The treatments with either sodium deoxycholate or acetone (Table 1, *a*–*e*) increased the total amount of phenolase activity compared with that in whole chloroplasts. The increases ranged from two and a half-fold with acetone (*c*) to six-fold with acetone and sodium deoxycholate (*e*). With the exception of treatment (*a*), all the extracts (*b*–*e*) had increased catecholase/cresolase ratios when compared with the original whole chloroplasts. The first treatment (*a*)

Table 1. SPECIFIC ACTIVITIES, TOTAL ACTIVITIES, DISTRIBUTION OF ACTIVITY AND INCREASE OF ACTIVITY RESULTING FROM EXTRACTION OF ISOLATED CHLOROPLASTS BY VARIOUS MEANS. SPECIFIC ACTIVITY IS MAXIMAL RATE OF OXIDATION OF SUBSTRATE MEASURED AS μL. OXYGEN/MGM. PROTEIN/MIN.

Experiment	Extraction treatment*	Fraction	Specific activity		Ratio Cat./Cres.	Total activity (catecholase) in fraction (specific activity × mgm. protein in fraction)	Percentage of total activity extracted into solution†	Increase in activity‡
			Catecholase	Cresolase				
a	1 per cent sodium deoxycholate, *p*H 7·3§, 30 min.	1 whole chloroplasts 2 extract 3 residue	1·13 4·9 6·0	1·13 4·9 5·5	1·0 1·0 1·09	34 108 46	70	× 4·5
b	1 per cent sodium deoxycholate, *p*H 6·3§, 10 min.; residue then treated with 1 per cent sodium deoxycholate, *p*H 7·3§ for 10 min.	1 whole chloroplasts 1*a* *p*H 6·3 extract 2 *p*H 7·3 extract 3 *p*H 7·3 residue	1·17 0 7·5 9·2	1·17 0 5·6 6·7	1·0 1·43 1·36	43 0 114 250	31	× 4·0 (allowing for totally inactive protein fraction 1*a*)
c	Acetone¶ followed by buffer *p*H 7·3, 10 min.	1 whole chloroplasts 2 extract 3 residue	2·25 7·4 3·5	1·9 5·2 2·6	1·17 1·42 1·32	53 70 64	52	× 2·5
d	Acetone¶ followed by buffer *p*H 8·0, 10 min.	1 whole chloroplasts 2 extract 3 residue	2·25 12·5 4·65	1·9 6·0 3·9	1·17 2·1 1·17	53 52 81	39	× 2·5
e	Acetone¶ followed by 1 per cent sodium deoxycholate, *p*H 7·3§, 10 min.	1 whole chloroplasts 2 extract 3 residue	2·25 29·2 8·5	1·9 17·0 6·45	1·17 1·7 1·32	53 162 155	51	× 6·0
f	1 per cent digitonin, *p*H 7·3§, 10 min.	1 whole chloroplasts 2 extract 3 residue	2·0 3·9 2·08	1·74 1·8 2·47	1·17 2·6 0·84	48 26·5 13·0	67	× 1·0 (that is, no activation)

* This shows the solution in which the chloroplasts were macerated, and the time during which they were left at 0° C. before centrifuging.

† $\dfrac{\text{Total activity in extract}}{\text{Total activity in extract + T.A. in residue}} \times 100$

‡ $\dfrac{\text{Total activity in extract + T.A. in residue}}{\text{Original total activity of whole chloroplasts}}$

§ Both the sodium deoxycholate and digitonin were dissolved in *M*/15 phosphate buffer at the *p*H's indicated above.

¶ Chloroplasts rapidly macerated with acetone and then centrifuged. Total time of contact therefore 5¼ min. Acetone residue then extracted as indicated above.

yielded the highest amount of activity in the extract (~ 70 per cent).

Digitonin (Table 1) also extracted about 70 per cent of the total phenolase, but in this case the extract had a greatly increased catecholase/cresolase ratio, and the residue a decreased one; furthermore, there was no increase in activity after the treatment. The activation of chloroplast phenolase by acetone and deoxycholate, on one hand, and the lack of activation by digitonin, on the other, may be comparable phenomena to those observed by Kenten[3] for the latent phenolase of bean leaf. He found that the latent phenolase is activated by anionic but not by non-ionic wetting agents.

It is important to note that with the treatments used, not all the phenolase could be extracted from the chloroplasts. This finding suggests that the enzyme is either tightly bound to, or forms part of, some structure in the sugar beet chloroplast. Phenolase may, therefore, have a specific function in the chloroplast, possibly associated with its high cresolase activity.

We thank Mr. G. E. Russell, Plant Breeding Institute, Trumpington, for generous supplies of sugar beet leaves and Dr. R. Hill and Dr. T. Swain for helpful discussions.

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[1] Arnon, D. I., *Plant Physiol.*, **24**, 1 (1949).
[2] Maver, A. M., and Friend, J., *J. Exp. Bot.* (in the press).
[3] Kenten, R. H., *Biochem. J.*, **67**, 300 (1957); **68**, 244 (1958).

Transmethylation in *Anodonta cygnea*

IN a previous survey of the distribution in Nature of betaine-homocysteine-methyl-transferases, these enzymes were found in the livers of all vertebrate animals investigated[1,2]. They could not be detected in any of a number of lower animals examined. The search for the enzymes has since continued, which has led to the discovery of a betaine–homocysteine–methyl-transferase in a member of the Mollusca, the pond mussel *Anodonta cygnea*. The work on which this observation is based will be briefly reported here.

The mussels were harvested in Lake Mälaren and transported in lake water to the laboratory. The livers were excised and homogenized in ice-cold phosphate buffer (0·05 *M*, *p*H 7·8) in a Potter–Elvehjem homogenizer. Methyl donors and homocysteine were added to the homogenate to give incubation mixtures having the compositions shown in Tables 1 and 2. Controls without additions, or containing only betaine or homocysteine, were also prepared. The amount of methionine formed after incubation at 37° C. for 2–4 hr. was estimated microbiologically[3] and in some instances also chemically[4].

Table 1. FORMATION OF METHIONINE BY HOMOGENATES OF *Anodonta* LIVER IN THE PRESENCE OF THE COMPOUNDS LISTED

Additions	Methionine formed (μgm./mgm. nitrogen per hr.)
None	1·8
0·025 *M* DL-homocysteine	2·9
0·1 *M* betaine	1·6
0·1 *M* betaine + 0·025 *M* homocysteine	8·3
0·1 *M* choline + 0·025 *M* homocysteine	3·0
0·1 *M* betaine + 0·025 *M* homocysteine + 0·1 *M* choline	8·3
0·1 *M* betaine + 0·025 *M* homocysteine + 0·1 *M* dimethylglycine	3·8

Table 2. EFFECT OF VARIOUS METHYL DONORS
All incubation mixtures contained 0·025 *M* DL-homocysteine

Methyl donor (0·05 *M*)	Relative activity (per cent)
Betaine	100
Betaine methyl ester	100
Betaine amide	0
Carboxyethyl-trimethyl-ammonium-chloride (propiobetaine)	0
Carboxypropyl-trimethyl-ammonium-chloride (butyrobetaine)	0
DL-Carnitine	0
1-Carboxyethyl-trimethyl-ammonium-chloride (α-methylbetaine)	45
Carboxymethyl-dimethylethyl-ammonium chloride	35
Carboxymethyl-methyldiethyl-ammonium chloride	5
Carboxymethyl-triethyl-ammonium chloride*	0*
Dimethylacetothetin†	380
Dimethylpropiothetin†	300
N-trimethyltaurine (taurobetaine)	0

* No ethionine was formed.

† No attempt was made to find optimal conditions for transmethylation from these compounds.

As can be seen from Table 1, the incubation mixtures containing betaine and homocysteine had a higher methionine content at the end of the incubation period than the controls. The presence of homocysteine alone consistently caused the formation of a small but significant amount of methionine, which suggests that the liver of *Anodonta* contains a donor of methyl groups. The addition of choline and homocysteine did not stimulate the synthesis of more methionine than did the addition of homocysteine alone. This indicates that choline is not in itself a methyl donor and that the choline oxidase activity of this tissue is low. The enzyme system causing the formation of methionine from betaine and homocysteine remained in the supernatant fluid after centrifuging homogenates prepared in 0·25 *M* sucrose at 105,000*g* for 60 min. Incubation of the supernatant fluid with ^{14}C-methyl betaine and homocysteine gave rise to radioactive compounds with the same R_F-values as methionine and dimethylglycine in three different solvent systems. The apparent *p*H-optimum of the reaction was near 8·5, which is higher than the optima observed for the methyl transferases of vertebrate livers.

The results in Table 1 also show that dimethylglycine, but not choline, inhibited the synthesis of methionine. Table 2 shows that the methyl donor requirement of the transferase of *Anodonta* liver could be satisfied to a greater or lesser extent by certain betaine derivatives and thetins. It is of interest to notice that the methyl ester of betaine possessed full donor activity, whereas the amide was completely inactive. The length of the carbon chain between the carboxyl group and the nitrogen is apparently of vital importance, as neither propio- nor butyro-betaine was able to serve as donor of methyl groups. Carnitine was also inactive. Both aceto- and propio-thetin, on the other hand, were excellent methyl donors. The introduction of a methyl group in the α-position relative to the carboxyl group in betaine decreased the activity, probably due to steric hindrance.

Replacement of one or two of the N-methyl groups of betaine with ethyl groups gave compounds having approximately 35 and 5 per cent of the activity of unsubstituted betaine. No evidence of ethyl group transfer from these compounds or from 'triethylbetaine' was obtained.

N-trimethyltaurine (taurobetaine), recently isolated from a species of Porifera by Ackermann *et al.*[5], was also tested as a methyl donor but did not seem to have this property. Nor did it act as an inhibitor of the enzyme.

Betaine–homocysteine–methyl-transferase activity could not be detected in any other organ than the liver of *Anodonta cygnea*.

I wish to thank Profs. F. Challenger, G. S. Fraenkel, D. Ackermann and Dr. B. Wickberg for supplying samples of acetothetin, carnitine, 'natural' taurobetaine and synthetic taurobetaine, respectively, and Mr. Nils Walde for help with the synthesis of the other compounds tested as methyl donors. The financial support of the Swedish Medical Research Council and the Knut and Alice Wallenberg Foundation is also gratefully acknowledged.

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[1] Ericson, L.-E., Fourth Int. Cong. Biochem., Vienna (1958), Abstracts of Communications, p. 45.

[2] Ericson, L.-E., *Acta Chem. Scand.*, 12, 1541 (1958), and to be published.

[3] Steele, B. F., Sauberlich, H. E., Reynolds, M. S., and Baumann, C. A., *J. Biol. Chem.*, 177, 533 (1949).

[4] McCarthy, T. E., and Sullivan, M. X., *J. Biol. Chem.*, 141, 871 (1941).

[5] Ackermann, D., and List, P. H., *Naturwiss.*, 10, 354 (1959).

Separation of the Hypnotic Potentiating Principles from the Essential Oil of *Acorus calamus* L. of Indian Origin by Liquid–Gas Chromatography

THE sedative potentiating activity of the essential oil of *Acorus calamus* L. of Indian and European origin has been reported[1].

It is the purpose of this communication to describe the isolation and identification of the active principle or principles responsible for the above-mentioned pharmacological activity from the essential oil of *Acorus calamus* L. of Indian origin.

In our investigations the volatile oil distilled from *Acorus calamus* L. of Indian origin by Fritzsche Bros., Inc., New York, was used, and liquid–gas chromatography was employed as a technique for the isolation of the individual components. The essential oil was first freed from phenols, acids and carboxylic compounds by the usual chemical procedures[2]. Subsequent to this the majority of other polar components were removed by passing the partly purified oil (75 gm.) through a silica-gel column (40 gm., 100–200 mesh, dried at 110° C.).

The purified oil (50–200 µl.), which exhibited the same pharmacological activity as the crude oil, was injected into an 'Aerograph' liquid–gas chromatography unit (Wilkens Instrument and Research, Inc., Walnut-Creek, California). The column consisted of a 10-ft. length of 1/4-in. stainless steel tubing packed with Johns–Mansville 'Chromosorb' (35–80 mesh) coated with 'Ucon' polar substrate in a ratio 75/25 by weight. The oven temperature was 205° C. and dried helium was used as carrier gas with a flow-rate of 64 ml. per min. measured at the outlet of the apparatus. These conditions were used throughout. Under these conditions the essential oil of *Acorus calamus* L. of Indian origin separated into a number of fractions (Fig. 1).

Four fractions were collected and tested pharmacologically. Fractions U_1 and U_2 appeared to be inactive pharmacologically. Fractions U_3 and U_4 potentiated the activity of pentabarbitone sodium. Fraction U_4 proved to be asarone, 1,2,4-trimethoxy-5-propenylbenzene (calculated C per cent 69·23; H per cent 7·69; found C per cent 69·13, H per

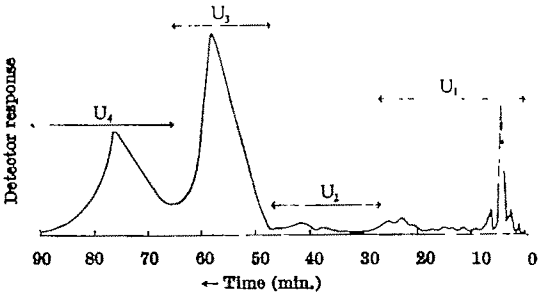

Fig. 1. Liquid-gas chromatogram of 200 μl. essential oil of *Acorus calamus* L. of Indian origin after removing the oxygenated compounds. Column, 'Ucon' polar on 'Chromosorb' 10-ft. × ¼ in. Temperature, 205° C. Flow, 64 ml. of helium per min.

cent 7·43). The compound melts at 58–61° C. (recorded m.p. 61° C.[3]): its picrate melts at 80° C. (recorded m.p. 81–82° C.[4]). Fraction U_3 is a colourless and odourless oil, which when treated with inorganic acids gives the same parasarone as fraction U_4 gives with similar treatment (m.p. 195–200° C., recorded m.p. for parasarone 202–203° C.[5]). After reduction with hydrogen in the presence of palladium charcoal a colourless oil was formed, the retention time of which, under the above-mentioned conditions, is identical with the retention time of the oil derived from fraction U_4 hydrogenated in the same manner. (The hydrogen consumed in both cases is about 1 mole calculated for trimethoxypropenylbenzene.) The ultra-violet spectra of fractions U_3 and U_4 are almost identical. (Absorbancy maxima occur at 253 mμ and 302 mμ.) From these results it can be concluded that the fraction U_3 and fraction U_4 are in *cis–trans* isomeric relationship, as was published earlier[6]—the fraction U_3 proved to be β-asarone.

It is to be noted that re-injection of fraction U_4, which was shown to be asarone, resulted in the appearance of two peaks on the chromatogram (U_4 and U_{4I}, Fig. 2, solid line). After several re-injections of the collected asarone, derived from the previous injection, fraction U_{4I} appeared repeatedly and the ratio between the two peaks did not change.

From these observations the conclusion may be drawn that the second peak is not due to a contamination but probably is due to a compound formed during the liquid–gas chromatography procedure. This assumption is supported by another experiment. When pure asarone was kept at 200° C. in a sealed tube under nitrogen for 24 hr., and the mixture formed injected into the column, two peaks, fraction U_4 and fraction U_{4I}, also appeared on the chromatogram, but the ratio between the peaks changed significantly compared to the former experiment when pure asarone was injected into the column. The quantity of fraction U_{4I} increased markedly (Fig. 2, dotted line).

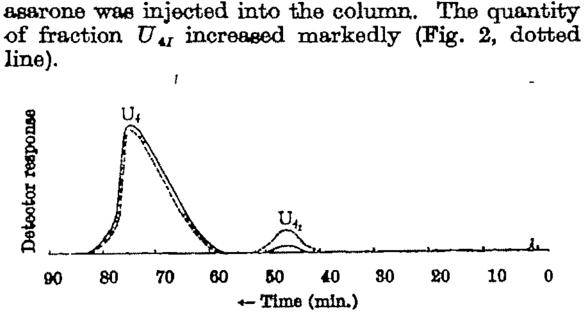

Fig. 2. Liquid–gas chromatogram of 100 μgm. U_4 (solid line) and 100 μgm. U_4 after keeping 24 hr. at 200° C. in a sealed tube under nitrogen (dotted line). Column, 'Ucon' polar on 'Chromosorb' 10 ft. × ¼ in. Temperature, 205° C. Flow, 64 ml. helium per min.

A small amount (about 150 μl.) of the fraction U_{4I} was collected and purified by liquid–gas chromatography. It is a light yellowish odourless oil, which does not crystallize even at temperatures of − 50° C. On treating with inorganic acid a colourless amorphous solid forms, which is different from parasarone. Its ultra-violet spectrum is almost identical with that of asarone and β-asarone. (Absorbancy maximum at 253 mμ and 302 mμ.) The pharmacological activity is different from asarone and β-asarone, in that it possesses not only hypnotic potentiation activity but also sedative activity.

Further studies are in progress to determine the chemical structure of this latter compound. Details of the pharmacological action and chemical nature of these compounds will be published elsewhere.

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[1] Dandiya, P. C., Baxter, R. M., Walker, G. C., and Cullumbine, H., *J. Pharm. Pharmacol.*, 11, 163 (1959).

[2] Kelkar, N. C., and Rao, B. Sanjiva, *J. Ind. Inst. Sci.*, 17, A, 7 (1933).

[3] Thoms, H., and Beckstroem, R., *Ber.*, 34, 1021 (1901).

[4] Bruni, G., and Tornani, E., *Atti accad. Lincei*, (5) 13, II, 186 (1904).

[5] Thoms, H., and Beckstroem, R., *Ber.*, 35, 3187 (1902).

[6] Rao, B. Sanjiva, and Subramaniam, K., *J. Chem. Soc.*, 1338 (1937).

Methylglyoxal Formation from Aminoacetone by Ox Plasma

IT was previously reported[1–3] that washed suspensions of *Staphylococcus aureus* synthesize aminoacetone from either threonine or glycine and the new aminoketone was isolated in crystalline form. Laver *et al.*[4] and Kikuchi *et al.*[5] have independently demonstrated the formation of aminoacetone by preparations of red blood cells and have shown that the system which forms δ-aminolævulinic acid is responsible for this activity.

I suggested recently[3] that aminoacetone might be involved in a metabolic cycle and postulated that it undergoes deamination to yield methylglyoxal. The present communication is concerned with this aspect of the metabolism of aminoacetone.

Preliminary studies showed that tissue slices of either rat liver or kidney catalysed a slow disappearance of the aminoketone, but so far it has not been possible to demonstrate an unequivocal enzymic attack on aminoacetone using cell-free extracts of these tissues. It was, however, observed that citrated ox plasma catalysed a rapid oxidation of aminoacetone to yield a product which, unlike aminoacetone, gave an immediate reaction with dinitrophenylhydrazine. The enzyme has been purified approximately tenfold by ammonium sulphate fractionation and this purified preparation was used for the subsequent work described here.

To identify the product of aminoacetone oxidation an incubation mixture containing 400 μmoles of aminoacetone (in a final volume of 120 ml.), plasma enzyme, crystalline catalase and phosphate buffer, *p*H 6·2, was shaken for 3 hr. at 37°. At the end of this period the aminoacetone had completely disap-

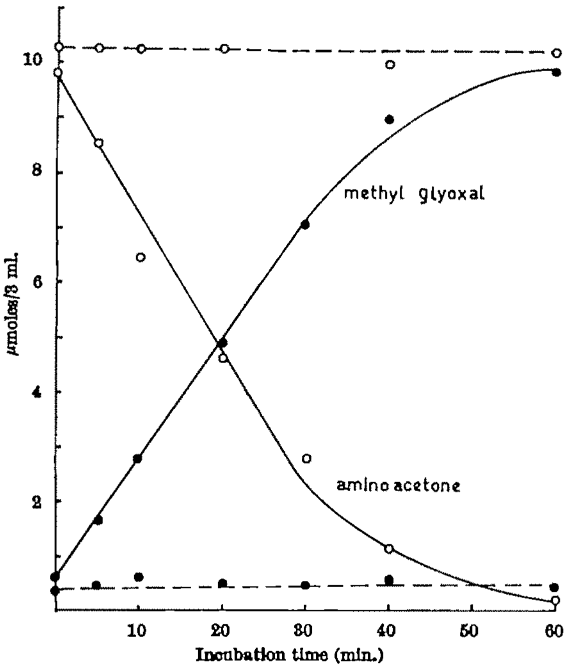

Fig. 1. Broken lines represent a control experiment in which the enzyme was boiled before addition

peared. After deproteinization the extract was passed through a column of 'Zeokarb 225' (*H* form) to remove any amino compounds, and the product isolated from the effluent as the dinitrophenylosazone. A control incubation mixture which was identical, except that aminoacetone was added after incubation, gave no osazone when treated in the same way. The product was recrystallized as orange-red needles from anisole. Yield: 0·056 gm., m.p. 298–300° (decomp.) (lit.[6] 295° decomp.; 299–300° decomp.). Found: C, 41·9; H, 2·9; N, 25·8. Calc. for methylglyoxal 2:4-dinitrophenylosazone, $C_{15}H_{12}N_8O_8$: C, 41·7; H, 2·8; N, 25·9 per cent. The isolated product gave the blue-violet colour in alkali characteristic of methylglyoxaldinitrophenylosazone. The identification of the product of aminoacetone oxidation as methyglyoxal was further confirmed by the observation that a partially purified ox heart glyoxalase preparation caused its disappearance, and this was dependent on the addition of glutathione.

Fig. 1 shows the progress of the enzymic reaction with time. Methylglyoxal was formed in amounts equivalent to the aminoacetone disappearing, and no reaction occurred when the enzyme was previously boiled. The aminoketone was quantitatively oxidized and no diminution in the reaction-rate took place until the substrate concentration was reduced to less than 10^{-3} *M*. In these experiments methylglyoxal was measured by a colorimetric method based on that described for pyruvate assay[7].

Manometric studies have shown that in the presence of added catalase, one atom of oxygen was taken up for each molecule of aminoacetone oxidized. In the absence of catalase, the initial rate of oxygen consumption was approximately double that in its presence, but the rate fell off rapidly. However, oxygen uptakes significantly in excess of one atom per molecule of aminoacetone present have been obtained under these conditions, indicating that the following reaction occurs:

$$CH_3.CO.CH_2NH_2 + O_2 \rightarrow CH_3.CO.CHO + H_2O_2 + NH_3$$

The purified enzyme used in these experiments contained amine oxidase activity[8] as measured by benzylamine oxidation and it is possible that aminoacetone oxidation is catalysed by this enzyme.

The present results are believed to be the first unequivocal demonstration of methylglyoxal formation from a substrate known to occur naturally and as such suggest a function for the glyoxalase enzyme system. The distribution of the aminoacetone oxidizing system in mammalian tissues is now being investigated.

Although all the steps of the postulated cycle[3] have now been shown to occur in suitable preparations, it nevertheless remains to be established whether the reactions take place as a metabolic pathway in living organisms. Experiments to this end are being undertaken.

I am indebted to Prof. A. H. Ennor for his stimulating interest in this work and for many helpful discussions.

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[1] Elliott, W. H., *Biochim. Biophys. Acta*, 29, 446 (1958).
[2] Elliott, W. H., *Nature*, 183, 1051 (1959).
[3] Elliott, W. H., *Biochem. J.* (in the press).
[4] Gibson, K. D., Laver, W. G., and Neuberger, A., *Biochem. J.*, 70, 71 (1958).
[5] Kikuchi, G., Kumar, A., and Shemin, D., *Fed. Proc.*, 18, 259 (1959).
[6] Rothstein, E., *J. Chem. Soc.*, 1553 (1940).
[7] Tonhazy, N. E., White, N. G., and Umbreit, W. W., *Arch. Biochem. Biophys.*, 28, 36 (1950).
[8] Tabor, C. W., Tabor, H., and Rosenthal, S. M., *J. Biol. Chem.*, 208, 645 (1954).

Evidence for the Hexose Monophosphate Pathway for Glucose Metabolism in Human Pancreatic β-Cells

THE level of glucose in the blood perfusing the islets of Langerhans is thought to regulate the secretion of insulin by the β-cells[1]. At present nothing is known about the mechanism of this regulation or even about the way in which the β-cells metabolize glucose. Studies designed to provide information about these problems are hampered by the fact that the pancreatic islets are rather diffusely scattered throughout the entire exocrine portion of the gland and comprise only about 2 per cent of the total pancreatic tissue[2]. Even if one were successful in obtaining islet tissue free of exocrine pancreas, the β-cells represent only 60–90 per cent of the total cell population[3]. However, in humans there are insulin-producing tumours of the pancreas which are usually composed of only β-cells and such tissue does afford an opportunity to study β-cell metabolism. A ratio greater than unity for the amount of carbon dioxide labelled with carbon-14 derived from glucose-1-^{14}C as compared to that from glucose-6-^{14}C has been taken as evidence for the existence of the hexose monophosphate pathway in the tissue under study[4]. The present report concerns studies using a β-cell tumour and indicates that there is an active hexose monophosphate pathway in these cells (this patient was studied through the kindness of the National Institute of Neurological Diseases and Blindness, National Institutes of Health, Bethesda, Md.). It is realized that the metabolism in these benign tumours may not be completely analogous to that of β-cells in normal islets.

Paired tissue slices weighing approximately 100 mgm. were incubated in glucose–Krebs Ringer bicarbonate buffer containing 0·5 μc. glucose-1-^{14}C or glucose-6-^{14}C and a final glucose concentration of 250 mgm. per cent. Incubations were done at 37° C. on a Dubnoff shaker under an atmosphere of 95 per cent oxygen and 5 per cent carbon dioxide for 90 min. In addition, some flasks also contained 0·01 mgm. of alloxan since this substance is known to destroy β-cells[5]. At the end of the incubation, hyamine was put in the centre well and the carbon dioxide was liberated from the incubation mixture by the addition of 10 *N* sulphuric acid. The hyamine containing labelled carbon dioxide was counted in a liquid scintillation counter using 0·4 per cent diphenyloxazole in toluene as the phosphor. Two control flasks without tissue but with 0·5 μc. glucose-1-^{14}C and glucose-6-^{14}C respectively were also included. For comparison similar experiments were done using rat hemidiaphragm, which is known to be devoid of the hexose monophosphate pathway[4].

The results are shown in Table 1. In slices from the β-cell tumour there were five to six times as many counts in labelled carbon dioxide when glucose-1-^{14}C was present as compared to glucose-6-^{14}C, indicating that the hexose monophosphate pathway was operative. In contrast, the radioactivity in labelled carbon dioxide from the rat muscle was approximately the same for both glucose-1-^{14}C and glucose-6-^{14}C. In addition, there did not appear to be any significant decrease in conversion of glucose to carbon dioxide in the presence of alloxan.

Table 1. CONVERSION OF GLUCOSE-1-^{14}C AND GLUCOSE-6-^{14}C TO $^{14}CO_2$ BY HUMAN PANCREATIC β-CELLS AND RAT HEMIDIAPHRAGM

Tissue	Counts/min./gm. in $^{14}CO_2$ from Glucose-1-^{14}C		Glucose-6-^{14}C	
β-cells	16,030	(2)	3,885	(2)
β-cells + alloxan	19,225	(2)	3,650	(2)
Rat hemidiaphragm	9,325	(2)	9,070	(2)

Number of determinations in parentheses

The full significance of the presence of an active hexose monophosphate pathway in β-cells cannot be ascertained although one can make some interesting inferences. Recently, the role of this pathway as a source of reduced triphosphopyridine nucleotide has been emphasized, and it has been suggested that it is of importance in many synthetic processes[6]. It has also been reported that stimulation of this pathway can lead to increased protein synthesis[7]. It is quite conceivable that the synthesis and secretion of insulin, which appears to be regulated in some manner by the blood glucose concentration, are thus facilitated by glucose metabolized via the hexose monophosphate pathway.

The finding that alloxan had no apparent effect on the conversion of glucose to carbon dioxide was somewhat unexpected. It may be that the dose of alloxan was too small or that alloxan, itself, is not toxic but that a metabolic product in the body is responsible for the β-cell damage. Other possibilities are that alloxan requires more than the 90-min. incubation period to disrupt β-cell metabolism or that the effects of this drug are not exerted on glucose catabolism but rather on some other essential cellular process[8]. It is also conceivable that β-cells in islet cell tumours are more resistant to alloxan than β-cells in normal islets.

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[1] Anderson, E., and Long, J. A., *Endocrinol.*, 40, 92 (1947).
[2] Hartroft, W. S., *Diabetes*, 5, 98 (1956).
[3] Volk, B., Lazarus, S., and Goldner, M., *Arch. Int. Med.*, 93, 87 (1954).
[4] Bloom, B., and Stetten, jun., D., *J. Amer. Chem. Soc.*, 75, 5446 (1953).
[5] Bailey, C. C., in "Vitamins and Hormones", 7, 365 (Academic Press, New York, 1949).
[7] Siperstein, M. D., *Diabetes*, 7, 181 (1958).
[8] Wilson, J. D., and Siperstein, M. D., *J. Clin. Invest.*, 38, 317 (1959).

ANIMAL PHYSIOLOGY

A Peanut Factor for Hæmostasis in Hæmophilia

IT is known that there are unpredictable apparent remissions of clinical symptoms enjoyed by hæmophiliacs; but these remissions have not been correlated with any influences such as time of year, food eaten, weather conditions, other diseases, or physical condition of the patient. The lack of 'antihæmophilic factor' associated with classic hæmophilia has been attributed[1] to a hypothetical block during the metabolic synthesis of the factor resulting from the action of the mutant gene of hæmophilia, such as occurs in the nutritional mutants of *Neurospora* and in eye pigment development in certain mutants of *Drosophila*[2]. In the cases of *Neurospora* and *Drosophila*, and other organisms studied, the addition of the substances the synthesis of which is blocked genetically results in normal metabolism. If this is the case in hæmophilia, it may be possible to supply, by mouth or injection, the unavailable material necessary for normal production of the hæmostatic substance lacking in hæmophiliacs. However, the few instances of success of various supplements, such as vitamins E and B_{12}, X-rays, and œstrogen, have not been confirmed[3-7].

One of us (H. B. B.) suffers from antihæmophilic factor hæmophilia, and in the past has been able to perform his duties with the help of periodic transfusions of fresh blood or infusions of lyophilized plasma following a hæmophilic attack. Various foods likely to overcome the hypothetic metabolic block by furnishing substances in the antihæmophilic factor pathway have been ingested in large daily quantities (blood pudding, liver, raw milk, eggs) without causing any reduction in severity of the disease. In May 1957, however, there seemed to have occurred a rapid loss, following consumption of a large handful of roasted peanuts, of tenderness (indicating a stoppage of bleeding) in a knee with an active hæmatoma. This loss in tenderness occurred without other therapy.

Since then, peanuts in all available forms (commercial peanut-butter, roasted, or raw peanuts) have been ingested on the appearance of each hæmophilic attack, and in each instance the clinical symptoms were relieved in one or two days.

During a period of more than two years, when peanuts, peanut flour, or a concentrate prepared from peanut flour has been in use, it was necessary only once to use lyophilized plasma to control a bleeding episode. This bleeding occurred in the lower pharynx, making ingestion impossible. Whenever peanut products are avoided, the tendency to bleed recurs, but yields to re-establishment of peanut regimen.

The relief of clinical symptoms brought about by peanut material is not correlated with reduction of capillary blood clotting time measured in glass capillary tubes, or of venous blood clotting time measured by the Lee–White method. No other attempts were made to measure any possible effects *in vitro*. In spite of the unreliability of the capillary tube method of measuring clotting time, the deepest punctures during periods of ingestion of peanut material have always healed without special treatment or rebleeding. The same normal healing has occurred in all cases of skin wounds occurring during the ingestion of peanut material.

Efforts have been made to concentrate the active factor. Peanut flour, prepared on extracting lightly roasted peanuts with hexane, and the crude extracted oil were each tested separately. The hæmostatic effect seems to be confined to the flour. It was found by Boudreaux, and by a second antihæmophilic factor hæmophiliac volunteer (aged fifty-three), that clinical symptoms are apparently relieved in one or two days with doses of 180 gm. of the flour per day. A third volunteer (aged five) has had relief from bleeding while taking flour or screw-pressed extracted meals. Boudreaux is at present effectively using screw-pressed extracted meal as a prophylactic.

The active factor is removed from peanut flour by extraction with 90 per cent ethyl alcohol. The residue was ineffective in relieving clinical symptoms, whereas the alcohol-extracted material was apparently effective in one or two days in doses of 14 gm. (the equivalent of 1 lb. of peanuts) per day, as indicated in tests carried out by Boudreaux and a fourth volunteer (aged eighteen). In the case of the fourth hæmophiliac, relief was obtained with the concentrate in two days, when he had failed to respond to 9 plasma infusions administered over a period of 3 weeks. A finger which was badly bruised in a motor-car door during the period of ingestion of peanut concentrate required no special treatment. During another bleeding episode, 12 infusions of fresh plasma given to this fourth volunteer in 10 days did not stop bleeding from the kidneys. Plasma was discontinued, and two days later the alcohol-extracted material was administered orally in 5-gm. quantities every 6 hr. No blood appeared in the urine on the second day. The volunteer was discharged from the hospital three days later.

It is unlikely that enough antihæmophilic factor was produced on ingestion of the concentrate to cause clinical relief, but enough to affect 'clotting time'. Enhanced thrombocyte action alone may be sufficient for hæmostasis without fibrin production[8]. Another possibility is that the peanut factor hastens capillary healing or improves capillary integrity without effect on blood hæmostatic reactions[9]. Current tests on hamsters in our laboratory suggest that vasoconstriction is enhanced by ingestion of the concentrate.

It is hoped that these limited observations will incite the interest of medical research to help determine the general occurrence of this phenomenon. Meanwhile, efforts are continuing to concentrate and identify the active factor, and to develop a bioassay on laboratory animals.

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[1] Brinkhous, K. M., and Graham, J. B., *Blood*, 9, 254 (1954).

[2] The literature is extensive on the nutritional mutants. A good summary of these phenomena may be found in "Genetics and Metabolism", Wagner, R. F., and Mitchell, H. K. (Wiley, New York, 1955).

[3] Kohl, H., *Deutsch. med. Wschr.*, 72, 279 (1947).

[4] Taubmann, G., *Deutsch. med. Wschr.*, 74, 1338 (1949).

[5] Prosperi, P., and Lottini, A., *Sperimentale*, 100, 258 (1950).

[6] Sotgiu, G., and Lenzi, G., *Haematologica*, 37, 321 (1953).

[7] Triebel, R., *Klin. Wschr.*, 22, 747 (1943).

[8] Zucker, M. B., *Amer. J. Physiol.*, 148, 275 (1947).

[9] Spaet, T. H., *Blood*, 7, 641 (1952).

Distribution of Tryptophan in the Brain

IT is well established that the central nervous system of vertebrates contains a substance which is biologically and chromatographically indistinguishable from 5-hydroxytryptamine. The tissues richest in 5-hydroxytryptamine are the diencephalon and mesencephalon, with little or none in the cerebellum or white matter. In the course of paper chromatographic studies on the distribution of 5-hydroxytryptamine in the brain of vertebrates, a second indole substance was noted and afterwards identified as the amino-acid, tryptophan. The distribution of tryptophan did not always follow closely that of 5-hydroxytryptamine, and this result may be of importance since it is believed that 5-hydroxytryptamine is formed by the decarboxylation of 5-hydroxytryptophan, an amino-acid formed from tryptophan.

Acetone extracts of tissues from freshly killed animals were subjected to paper chromatography in four solvents: (1) 8 per cent (w/v) sodium chloride solution, (2) *iso*propanol/ammonia/water (20 : 1 : 2), (3) *n*-propanol/water (3 : 1), (4) *n*-butanol/acetic acid/water (4 : 1 : 5). The identity of tryptophan was established chemically by its R_F values in the different solvents and by its colour reactions with ninhydrin, Ehrlich's reagent, Pauly's reagent and *N.N.C.D.* reagent. Further, superposition of the tryptophan spot given by the tissue extracts and that given by tryptophan added to such extracts was obtained and a similarity of the colour reactions was observed.

The brain of dog, cat, mouse, guinea pig, rat, rabbit, pigeon, hen and gull contained between 1 and 3 μgm. tryptophan per gm. of fresh tissue, and these concentrations were between 3 and 10 times the corresponding concentrations of 5-hydroxytryptamine. Other tissues in the body such as the spleen, gut and blood also contained 3–10 times as much tryptophan as 5-hydroxytryptamine.

When the various parts of the brain of the dog, cat and rabbit were dissected out and assayed for their tryptophan content, the richest sources were found to be the pons and hypothalamus. The mid-brain medulla and cerebellum contained appreciable quantities whereas the cerebral hemispheres and spinal

Table 1. ESTIMATES OF TRYPTOPHAN AND 5-HYDROXYTRYPTAMINE (µGM /GM. TISSUE) IN DOG BRAIN

Part of brain	Tryptophan	5-Hydroxytryptamine
Cerebral hemispheres	0 1	0·01
Hypothalamus	5–40	0 40
Mid-brain	2–8	0 35
Cerebellum	1–4	0 02
Pons	1–20	0 05
Medulla	2–5	0·25
Spinal cord	0·2	0·02

cord showed only traces. The results for the dog are shown in Table 1 together with estimates of 5-hydroxytryptamine for the corresponding areas. Inspection of the figures shows that the tryptophan content of three areas—the hypothalamus, cerebellum and pons—was about 100 times that of 5-hydroxytryptamine whereas the ratio was only 3–10 for the other parts. A similar result has also been found for cat and rabbit brain. The reason why the hypothalamus, cerebellum and pons are relatively rich in tryptophan and poor in 5-hydroxytryptamine is not clear at present.

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Resting Potentials in Isolated Frog Sartorius Fibres at Low External Potassium Concentrations

IN earlier experiments referred to by Conway[1,2], it was shown that the resting potential (E) of isolated frog sartorii fibres corresponded to the theoretical values as determined by the relation:

$$E = \frac{RT}{F} \ln [\mathrm{K}]_o/[\mathrm{K}]_i = \frac{RT}{F} \ln [\mathrm{Cl}]_i/[\mathrm{Cl}]_o \qquad (1)$$

provided the conditions were such that a 'balanced state' (state of zero net flux) existed for the sodium and potassium ions.

The validity of the Conway theory, from which such an equation may be derived, has been well tried and supported by Kusel and Netter[3], by Mond[4], Adrian[5], Hodgkin[6]; Hodgkin and Horowitz[7]; also for mammalian skeletal muscle by Pillat, Kraupp, Giebisch and Stormann[8].

Here various potential measurements using the micro-electrode technique[9] were carried out[10] with fibres of frog sartorii which had been immersed overnight at 0–3° C. in Ringer–Conway fluid with constant sodium concentration, and potassium values up to very high concentrations, as great as 200 mM [11].

A series of determinations was also carried out with the impermeant anion, acetate, replacing all the chloride in the external solution, potassium being substituted for sodium up to high levels, the solution being thus maintained isotonic.

In both these types of experiment the theoretical potential with respect to the potassium ion was obtained above external values greater than 10 mM. Below this level the potential tended to fall below the theoretical figure, corresponding with the observations of Adrian, and of Hodgkin and Horowitz.

Immediate determinations of the resting potential of isolated fibres using Ringer–Conway fluid with normal plasma potassium level of 2·5 mM gave a value of 92·5 ± 0·47 mV., the theoretical figure being 100·1 mV. (the 'fibre water' potassium was 133 mM for the group of frogs used here). Adrian obtained similar results, the average fibre water potassium of his frogs being 139 mM.

The points on the curve of log [K] against E were shown by Hodgkin and Horowitz to be reasonably well fitted at the low potassium levels of 1–10 mM and 20° C. by the relation:

$$E = 58 \log \frac{[\mathrm{K}]_o + 0{\cdot}01\,[\mathrm{Na}]_o}{[\mathrm{K}]_i + 0{\cdot}01\,[\mathrm{Na}]_i} \qquad (2)$$

Above an external concentration of potassium of 10 mM, this equation gives results practically identical with equation 1. The fall below the potential-levels from equation 1 at external values of potassium less than 10 mM could be assigned initially to either of two causes. First, to the existence of a sodium–potassium linked pump which would result in an equation of the second type given above.

It could also arise if a 'balanced state' with respect to sodium and potassium ions did not in fact exist for isolated sartorii fibres with the external potassium concentration less than 10 mM. These two views are also referred to by Hodgkin and Horowitz[7], with the conclusion that their published observations are consistent with either of them.

To examine the question here the observation of Carey and Conway[12] was utilized that with plasma as the external medium the permeability of the fibres to sodium ions was retained closer to the normal.

Plasma was prepared by bleeding through an aortic cannula into a heparinized tube. The blood was centrifuged at once and the plasma analysed for its potassium content, suitably diluting or adding potassium chloride if necessary to give 2 5 mM.

Table 1

External fluid	External potassium concentration	Resting potential observed mV.	Resting potential theoretical mV.
Ringer–Conway	2·5	92 5 ± 0·47	100·1
Plasma	2 5	99·2 ± 0·7	100 1

Such results show that the hypothesis of a sodium–potassium linked pump is unnecessary with respect to the potential measurements for frog skeletal muscle, and is not the explanation of the fall from the theoretical potential values from equation 1, for isolated muscle at low external potassium-levels.

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[1] Conway, E. J., "Metabolic Aspects of Transport Across Cell Membranes", ed. Q. R. Murphy (The University of Wisconsin Press, 1957).

[2] Conway, E. J., *Physiol. Rev.*, **37**, 84 (1957).

[3] Kusel, H., and Netter, H., *Biochem. Z.*, **323**, 39 (1952).

[4] Mond, R., *Pflugers Archiv. ges. Physiol.*, **261**, 243 (1955).

[5] Adrian, R. A., *J. Physiol.*, **133**, 631 (1956).

[6] Hodgkin, A. L., *Proc. Roy. Soc.*, B, **148**, 1 (1957).

[7] Hodgkin, A. L., and Horowitz, P., *J. Physiol.*, **148**, 127 (1959).

[8] Pillat, B., Kraupp, O., Giebisch, G., and Stormann, H., *Pflugers Arch. ges. Physiol.*, **266**, 459 (1957).

[9] Graham, J., and Gerard, R. W., *J. Cell. Comp. Physiol.*, **29**, 99 (1946)

[10] Kernan, R. P., and Conway, E. J., Abstr. Int. Congr. Biochem (1955).

[11] Boyle, P. J., and Conway, E. J., *J. Physiol.*, **100**, 1 (1941).

[12] Carey, M. J., and Conway, E. J., *J. Physiol.*, **125**, 232 (1954).

Neutralization of Human Serum and Pituitary Thyrotropic Fractions with Antisera to Bovine Pituitary Thyrotropin

ANTISERA to pituitary growth hormone have been shown to possess a high degree of species specificity[1]. This was demonstrated by precipitin and by neutralization experiments, in which the antiserum to human growth hormone was active against human and monkey growth hormone, but not against growth hormone derived from other species.

Experience over the past years with antisera to a thyrotropic preparation of beef pituitary has revealed that the antisera do not possess great specificity when tested against thyrotropin from several other species[2]. In the present work, the observations have been extended to man. Antibovine thyrotropin sera have been found to neutralize the thyrotropic activity of fractions prepared from human pituitaries and from two individual sera, one from a patient with classical Graves's disease and the other from a healthy euthyroid subject.

Rabbits were immunized by the method of Freund[3], using a commercial bovine thyrotropin preparation in current clinical use ('Thytropar') as the antigen. Details of the methods employed and the results from precipitin and neutralization studies with the parent and other more purified thyrotropic extracts are being published elsewhere. The antibody-containing globulin fraction was concentrated from the antisera and from normal rabbit globulin by sodium sulphate precipitation[4].

The serum fractions were prepared by the method of Bates *et al.*[5]. The human pituitary preparation, not at the final stage of purification, was made by Heideman and Bakke[6] from pooled glands obtained at autopsy and was obtained through their courtesy. The active material contained 10 mμ/c.c.

The hormone to be neutralized and normal serum or antiserum, or the globulin preparation made from antiserum or normal serum, were mixed and kept overnight at 4° C. In one experiment (Exp. 234), previous absorption of both normal and antiserum with bovine serum was conducted prior to mixing. Assay for thyrotropic activity was performed by a slight modification of the method of McKenzie[7]. The iodine-131 content of the blood was measured before and 2 hr. after intravenous injection of the test sample. In each assay, saline, the human pituitary or serum preparation mixed as above with serum or globulin preparation of serum, and two dose-levels of a standard preparation of beef thyrotropin were assayed concurrently. Four mice were employed for each group.

It is evident from Table 1 that there was complete neutralization of human thyrotropin from both pituitary (Exps. 138, 230 and 234) and serum (Exps. 208 and 188) sources by the several anti-bovine thyrotropin sera used. No neutralization occurred when normal rabbit serum was employed. In Exp. 230, the

Table 1. NEUTRALIZATION OF THYROTROPIN FROM HUMAN PITUITARY AND SERUM BY ANTI-BEEF THYROTROPIN SERUM

Exp. No.	Test material	No. animals	Dose		Mouse assay (per cent of initial blood radioactivity at 2 hr.) Mean ± *S.D.*	*P* value*	Remarks
			TSH	Serum			
138 and 140	Saline	10			97 ± 26		Antiserum to relatively crude bovine thyrotropin
	Human TSH	10	0·5μ		267 ± 133		
	Human TSH + normal rabbit serum globulin	6	0·5μ	3·0 mgm. (0·15 c.c. equiv.)	337 ± 101		
	Human TSH + antiserum globulin	4	0·5μ	2·0 mgm. (0·1 c.c. equiv.)	113 ± 44	*P* < 0·001	
230	Saline	4			68 ± 19		Antiserum to highly purified bovine thyrotropin, 25 μ/mgm. (ref. 10)† Pre-immune serum obtained from same rabbits as post-immune
	Human TSH	4	0·5μ		278 ± 90		
	Human TSH + pre-immune rabbit serum	4	0·5μ	0·5 c.c.	219 ± 51		
	Human TSH + post-immune rabbit serum	4 4	0·5μ	0·5 c.c.	106 ± 31	*P* < 0·001	
234	Saline	4			81 ± 11		Antiserum to highly purified bovine thyrotropin, 25 μ/mgm. (ref. 10)† Absorption of both normal and anti-serum with bovine serum before experiment
	Human TSH	4	0·25μ		170 ± 35		
	Human TSH + normal rabbit serum	4	0·25μ	0·5 c.c.	147 + 44		
	Human TSH + antiserum	4	0·25μ	0·5 c.c.	73 ± 11	*P* < 0·001	
208	Saline	4			94 ± 16		Antiserum to relatively crude bovine thyrotropin; absorption with human hypopituitary serum before experiment
	Bovine TSH	4	0·1μ		325 ± 36		
	Toxic diffuse goitre serum preparation‡	4	7·5 mgm. (1 c.c. equiv.)		297 ± 64		
	Toxic diffuse goitre serum preparation + antiserum globulin	4	7·5 mgm. (1 c.c. equiv.)	3·1 mgm. (0·4 c.c. equiv.)	92 ± 18	*P* < 0·001	
188	Saline	4			129 ± 50		Antiserum to relatively crude bovine thyrotropin
	Human TSH	4	0·1μ		240 ± 39		
	Normal human serum preparation	4	0·05 mgm. (0·01 c.c. equiv.)		284 ± 61		
	Normal human serum preparation + normal rabbit serum globulin	4	0·05 mgm. (0·01 c.c. equiv.)	10·0 mgm. (1 c.c. equiv.)	227 ± 30		
	Normal human serum preparation + antiserum globulin	4	0·05 mgm. (0·01 c.c. equiv.)	10·0 mgm. (1 c.c. equiv.)	153 ± 26	*P* < 0·001	

* *P* values calculated for difference between the means of values for thyrotropin preparation plus normal serum or antiserum, except in experiment 208, where difference was calculated from test serum fraction alone.

† Experiment confirmed with an antiserum to a preparation containing 25 μ/mgm. kindly provided by Dr. John G. Pierce, University of California.

‡ Insufficient preparation for test with normal rabbit serum globulin.

normal serum was obtained from the same rabbit in which the antiserum was induced. In Exp. 234, the normal serum was absorbed with normal bovine serum in the same manner as the antiserum. The failure of the bovine serum absorbed antiserum to neutralize the thyrotropin excludes the possibility of precipitation of hormonal activity along with non-specific proteins. The failure of the pre-immune and normal rabbit sera to neutralize excludes the presence of neutralizing substances in serum prior to immunization.

The nature of the material in human serum which induces a thyrotropic response, especially in Graves's disease, has not been as yet clearly identified as thyrotropin. Adams and Purves have observed that the release of iodine-131 from the thyroid is delayed following injection of thyrotoxic serum compared to standard preparations or to hypothyroid serum[8]. They suggested that possibly the thyrotropin in the serum of Graves's disease patients may be abnormal. This observation in the guinea pig has been confirmed in the mouse[7].

More recent work by McKenzie indicates that the material in hyperthyroid sera will cause histological activation of the mouse thyroid, an increased uptake of iodine-131, and an increase in level of protein-bound iodine[9]. This suggests that the active material in the serum is a thyrotropin. The delay before activation of the thyroid, however, by such a sera could also imply that the serum factor is not a thyrotropin, but only acts indirectly to cause the mouse to release its own thyrotropin.

As stated, the present experiments reveal that the thyrotropic effect of the thyroid-activating substances in normal and in hyperthyroid serum were equally well neutralized by an antithyrotropic serum. This suggests that the active material in both is a thyrotropin which is closely similar if not identical to that from the pituitary. It is not proved that endogenous mouse thyrotropin was not secreted and then neutralized by the injected antiserum. However, the extent of the dilution of the antiserum in the circulation of the mouse makes this possibility less likely, as does the short time, 2 hr., between injection and evidence of thyroid response. This allows only a very brief interval for the diluted antibody to have reacted with endogenous thyrotropin were it secreted.

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[1] Hayashida, T., and Li, C. H., Paper presented at the forty-first meeting of the Endocrine Society (June 4–6, 1959).

[2] Werner, S. C., *Endocrinol.*, **22**, 291 (1938).

[3] Freund, J., Lipton, M. M., and Morrison, L. R., *Arch. Path.*, **50**, 108 (1950).

[4] Howe, P. E., *J. Biol. Chem.*, **49**, 93 (1921).

[5] Bates, R. W., Garrison, M. M., and Howard, T. B., *Endocrinol.*, **65**, 7 (1959).

[6] Heideman, M. L., Bakke, J. L., and Lawrence, N. L., *J. Clin. Endocrinol. and Metab.*, **19**, 831 (1959).

[7] McKenzie, J. M., *Endocrinol.*, **62**, 865 (1958).

[8] Adams, D. D., and Purves, H. D., *Metabolism*, **6**, 26 (1957).

[9] McKenzie, J. M., *J. Clin. Endocrinol. and Metab.* (in the press).

[10] Condliffe, P. G., and Bates, R. W., *Arch. Biochem. and Biophys.*, **68**, 229 (1959).

A Substance in Lingcod Liver Oil which prevents Hypercholesterolæmia in Cholesterol-fed Chickens

THE serum cholesterol-level in some animals is elevated above the normal value by the addition of cholesterol to the diet. In certain species, such as the chicken, cholesterol by itself brings about this increase ; but in other species, exemplified by the rat, the presence of cholic acid is also required. The degree of hypercholesterolæmia is altered when certain vegetable and fish oils are added to the diet ; but the observed effect of any one oil is dependent on the species of animal under investigation. De Groot and Reed[1] showed that cod liver oil and corn oil prevented the increase in plasma cholesterol in rats due to dietary cholesterol and cholic acid. Dam *et al.*[2] investigated the effect of dietary cod liver oil and linseed oil in cholesterol-fed chickens, and found that both oils increased the hypercholesterolæmia caused by the dietary cholesterol. Similar experiments on chickens were performed by Wood and Biely[3] using corn oil, tallow and various fish oils. Corn oil, tallow and herring oil were observed to increase the hypercholesterolæmia ; but lingcod liver oil and halibut liver oil completely prevented the appearance of hypercholesterolæmia. Ratfish liver oil, dogfish liver oil and basking shark liver oil were intermediate in effect. In view of the diverse effects of the oils observed in the chicken studies, further investigations were carried out to obtain additional information about the mode of action of the oils. Corn oil, herring oil and lingcod liver oil were chosen for the investigation.

The oils were saponified with ethanolic potassium hydroxide, and the soap solutions were extracted several times with ethyl ether to obtain the unsaponifiable material. The soap solutions were then acidified with hydrochloric acid and the free fatty acids extracted with ethyl ether. The extracts containing the unsaponifiable fractions and the free fatty acids were washed and the solvent removed on a water-bath at 70° C. and under a stream of nitrogen. The oils and their components were added to a diet containing 1 per cent cholesterol. This diet was fed to 2-weeks old New Hampshire chickens for 9 days, after which time blood samples were taken and the serum cholesterol concentrations determined. The oils were fed at a 10 per cent level in the diet and the unsaponifiable substances and the free fatty acids were added in amounts approximating to that obtained from an equivalent amount of oil.

As observed previously, lingcod liver oil prevented the hypercholesterolæmia caused by the dietary cholesterol, and herring oil and corn oil stimulated the rise in serum cholesterol concentration (Table 1). The free fatty acids from all three oils markedly increased the degree of hypercholesterolæmia, whereas

Table 1. EFFECT OF VARIOUS OILS AND FRACTIONS FROM THESE OILS ON SERUM CHOLESTEROL-LEVELS IN CHOLESTEROL-FED CHICKENS

Addition to the cholesterol-containing diet	Serum cholesterol (mgm./100 ml.)
None	192 ± 12*
Corn oil	784 ± 173
Herring oil	648 ± 133
Lingcod liver oil	130 ± 31
Corn oil fatty acids	793 ± 144
Herring oil fatty acids	618 ± 215
Lingcod liver oil fatty acids	702 ± 158
Corn oil unsaponifiables	193 ± 21
Herring oil unsaponifiables	237 ± 34
Lingcod liver oil unsaponifiables	119 ± 17

* Each value is the mean for four samples of serum ± the standard deviation. Each sample contained the serum from four chickens.

the unsaponifiable material from herring oil and corn oil produced only a slight increase or none at all. The unsaponifiable portion of lingcod liver oil, however, completely prevented the rise in serum cholesterol-level.

It was concluded that the ability of lingcod liver oil to prevent hypercholesterolæmia in cholesterol-fed chickens depends on the presence of some compound or compounds in the unsaponifiable material of the oil. At the present moment the active material has not been identified; but the possibility exists that it may be vitamin A or perhaps a sterol-type compound which acts in a similar manner to cholestanol. The latter compound was shown[4] to prevent effectively the appearance of hypercholesterolæmia induced in chickens by cholesterol feeding.

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[1] De Groot, A. P., and Reed, S. A., *Nature*, **183**, 1191 (1959).

[2] Dam, H., Kristensen, G., Nielsen, G. F., and Søndergaard, E., *Acta physiol. Scand.*, **45**, 31 (1959).

[3] Wood, J. D., and Biely, J., *Can. J. Biochem. and Physiol.* (in the press).

[4] Siperstein, M. D., Nichols, jun., G. W., and Chaikoff, I., *Circulation*, **7**, 37 (1953).

SEROLOGY

A Starch Gel Immunoelectrophoretic Analysis of Human γ-Globulin demonstrating the Inter- and Intra-Specific Variation of Organisms

AN age-dependent, phenotypic variation in the antigenic composition of human γ_2-globulin has been reported[1-3]. Chicken antisera to preparations rich in γ-globulin (for example, *FrII*) were used to examine human sera by the starch gel immunoelectrophoretic method of Poulik[4] as modified by Goodman[3,5]. Certain of these antisera detected in many instances more than one component in that portion of the γ-globulin which moves backward from the origin in starch gel electrophoresis, that is, in the γ_2 fraction of serum. Recently, as the point of departure for the present work, non-human primate sera were also examined by some of the antisera. It was reasoned that, since the evolutionary continuum leads from the intra- to the inter-specific variation of organisms, the serum antigenic specificities which differ between human beings would fail to have derivative counterparts in the sera of the non-human primates. A considerable body of data illustrating the applications of this reasoning has now been gathered, and several examples of the data are presented in Figs. 1 and 2.

The immunoelectrophoretic reactions depicted in Fig. 1 show that the absorption of chicken antiserum with rhesus monkey serum selectively lowers the antibody concentration to the predominant antigenic groupings of human γ-globulin and thus permits a visualization of those groupings held by only a fraction of the γ_2-globulin molecules, a fraction which varies markedly between human beings, and which appears to occur more richly in adult sera than in prepuberty sera[2,3]. Note that such a fraction could be detected in the 'positive' human serum pool (made up of several adult sera) after antisera *C*-341 and *C*-342 were absorbed with the rhesus serum. The fragmented γ_2-globulin precipitin arc signifying a positive reaction was observed with single serum specimens as well as with the 'positive' pool. (An example of such a result is photographically reproduced in Fig. 1*E*.) On the other hand, a large number of the serum specimens from adults and children, as well as the 'negative' pool (made up of several prepuberty sera), failed to yield the second γ_2 reaction with these two reagents.

The results obtained with the antisera, *C*-79 and *C*-82, present a somewhat different picture, for these antisera each contained, as compared to *C*-341 and *C*-432, antibody directed to a large percentage of the spectrum of antigenic groupings showing an inter-specific variation between man and monkey. The additional γ_2 arc developed with the 'negative' pool as well as with the 'positive' pool. However, Fig. 1 (*C* II and *D* II) does show that the absorption with the rhesus serum left a more potent reaction for the variant γ_2 fraction of the 'positive' pool than for that of the 'negative' pool.

A number of human and non-human primate sera have now been surveyed for the presence or absence of the additional γ_2 reaction by unabsorbed *C*-79 and by *C*-82 after an absorption with a small quantity

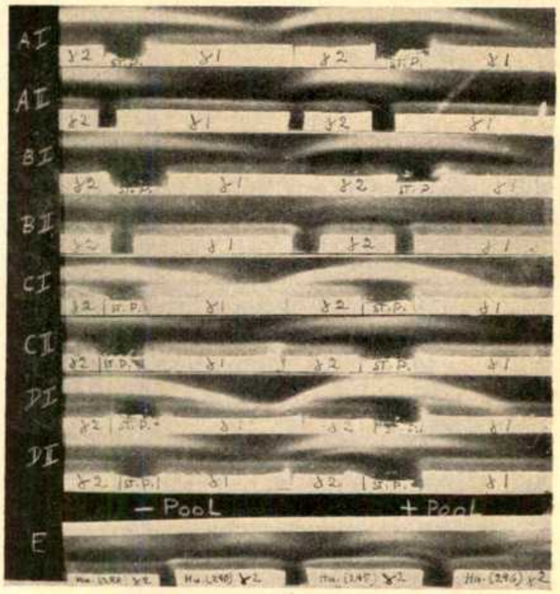

Fig. 1. Photographs of the reactions by double diffusion in agar gel of chicken anti-human γ-globulin sera against human serum separated by starch-gel electrophoresis. From *A* to *D* each reaction tray contains at its bottom the γ-globulin portions from the starch-gel separation of a 'negative' human serum pool and a 'positive' human serum pool (see text). Four different antisera are represented: *C*-341 in *B*, *C*-82 in *C* and *C*-79 in *D*. In each instance the reactions of the antiserum before absorption (I) are compared to those obtained after an absorption (II) with an aliquot of rhesus monkey serum estimated to contain 198 mcgm. γ-globulin per ml. of antiserum. In *E* the rhesus absorbed *C*-341 reacts against the γ_2-globulin portions of four different adult human sera. The second γ_2-precipitin arc is marked in samples 295 and 296 but weak to negative in samples 288 and 290. The non-specific lateral band that develops in the agar near the surface of the starch gel block can be so recognized and thus discounted in assessing the specific antibody–antigen reactions. γ_2, The γ_2-globulin portion of the starch gel block; *st.p.*, starch paste, in which the serum sample was originally inserted: γ_1, the γ_1-globulin of the starch gel block

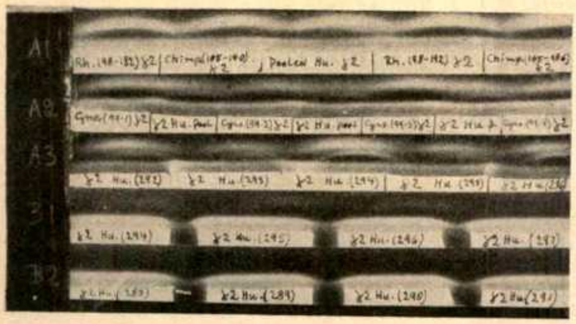

Fig. 2. Photographs of immunoelectrophoretic results obtained on surveying sera from the rhesus monkey (*Rh*), the cynomolgus monkey (*Cyno*), the chimpanzee (*Chimp*), and the human (*Hu*) for the presence or absence of a 'minor' γ_2-globulin antigen as detected by absorbed antiserum *C*-82 reacting against the γ_2-globulin portion (γ_2) of the starch gel blocks. For the *A* trays, *C*-82 was absorbed per ml. with an aliquot of 'negative' human serum estimated to contain 125 mcgm. of γ-globulin. A 'minor' γ_2-reaction occurring underneath the 'major' precipitin reaction can be seen only for the human samples. In the *B* trays, *C*-82 was absorbed per ml. with 410 mcgm. of γ-globulin of the appropriate human serum. In the two trays represented positive reactions can only be seen for human sera 294, 295 and 296. It is of interest that 294, 295, 296 and 297 are *Gm*-like (−) samples and 288, 289, 290 and 291 are *Gm*-like (+) samples. (The absorbed *C*-82, however, was not the best reagent of those tried for distinguishing *Gm*-like (−) samples from *Gm*-like (+) samples.)

of the 'negative' human serum pool which by weakening the predominant precipitin arc allowed the additional γ_2 arc to be fully revealed. All the adult human sera tested (14 samples with *C*-79 and 39 with *C*-82) yielded this additional reaction, though to varying degrees, but none of the non-human primate sera did. The survey has encompassed sera from 13 chimpanzees, a gibbon, 16 rhesus monkeys, 15 cynomolgus monkeys, 2 baboons, 2 mandrills, a langur, 2 spider monkeys, and a capuchin monkey. Examples of the results obtained in the survey are presented in Fig. 2*A*. Also note (Fig. 2*B*) that when *C*-82 was so absorbed with the appropriate serum pool that it could no longer yield in immunoelectrophoresis its 'major' γ-globulin precipitin arc, its 'minor' γ_2-precipitin arc (the one missing from non-human primates) was no longer developed by all human adult sera, but only by some.

The relationship of the antigenic components detected by such chicken antiserum reagents to the *Gm* groups[6] is now under investigation. For this purpose Dr. Grubb has kindly furnished 6 *Gm* (*a* +) sera and 6 *Gm* (*a* −) sera. Similarly, Dr. Steinberg has kindly furnished 6 of his *Gm*-like (+) sera which are present in some Negroes and rare or absent in whites, and 6 of his *Gm*-like (−) sera[7]. Although none of the reagents has distinguished the *Gm* (*a* +) group from the *Gm* (a −) group (each yielding positive and negative reactions with both groups), some of these reagents did distinguish the *Gm*-like (+) group from the *Gm*-like (−) group in that they reacted only with *Gm*-like (−) samples and not with the *Gm*-like (+) samples. The interrelationships of such γ-globulin specificities may prove to be so complex as to argue for the thought that genetic information inherited through a number of gene loci rather than just through a *Gm* locus[8] goes into shaping the antigenic architecture of a γ_2-globulin molecule. In this regard, an answer is being sought to the crucial question of whether there is in addition to the variant antigenic configurations detected an overall antigenic resemblance between the above 'major' and 'minor' fractions of the γ_2-globulin of human serum.

This investigation was aided in part by research grants from the National Institutes of Mental Health and the (American) National Multiple Sclerosis Society. I am indebted to Dr. Albert B. Sabin at the Children's Hospital Research Foundation in Cincinnati, and to Dr. Sidney Gains, chief of the Department of Microbiology at the Walter Reed Army Institute of Research, for providing me with the chimpanzee serum, and to Dr. G. D. Brigham, director of the Biological Division at Parke, Davis and Co., for the rhesus and cynomolgus monkey sera. I am also grateful to Dr. George B. Rabb, curator of the Chicago Zoological Park, for the many samples of primate sera. The expert assistance of Emily Poulik, Roger Gudobba, and Rosemarie McBride is greatly appreciated.

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[1] Goodman, M., Ramsey, D. S., and Poulik, E., *Fed. Proc.*, **17**, 514 (1958).

[2] Goodman, M., and Schwimmer, B., *Fed. Proc.*, **18**, 570 (1959).

[3] Goodman, M., *Amer. Nat.* (in the press).

[4] Poulik, M. D., *J. Immunol.*, **82**, 502 (1959).

[5] Goodman, M., Newman, H. S., and Ramsey, D. S., *J. Lab. and Clin. Med.*, **51**, 814 (1958).

[6] Grubb, R., *Acta Path. Microbiol. Scand.*, **39**, 145 (1956).

[7] Steinberg, A. G., Giles, B. D., and Stauffer, R., *Amer. J. Human Genet.* (in the press).

[8] Grubb, R., and Laurell, A. B., *Acta Path. Microbiol. Scand.*, **39**, 390 (1956).

Changes in Outdated Human γ-Globulin Preparations

THE fate of antibodies in normal human γ-globulin preparations during storage was studied by Gerlough[1], who succeeded in achieving surprising results: the half-time of diphtheria antitoxin in the preparation was 23 years—under strict storage conditions even 40–69 years. Tiselius electrophoresis and ultracentrifugation did not reveal cleavage or denaturation of the protein molecule.

We have studied a number of outdated normal human (plasmatic) γ-globulin preparations prepared by Cohn's classical methods 6 and 9 as well as by other methods. The products were stored at + 4° C. for 3–6 years in the form of sterile solutions. No difference was observed in the contents of diphtheria antitoxin compared with fresh products, though immunoelectrophoretically two atypical fractions besides the normal precipitation line were noticed. Of these atypical fractions one possessed a mobility lower (called γ_X) and the other higher (γ_Y) than γ-globulin. The γ_Y component appears sooner than the γ_X one, so that some of the preparations studied possessed only the γ_Y component and some both of them. Both fractions are not precipitated by antisera with saturated anti-γ-globulin antibodies. Fig. 1 presents two γ-globulin products prepared four years ago by Cohn's classical method. These products were originally absolutely homogeneous. When studying the γ-globulins prepared from materials containing hæm and hæmoglobin a similar phenomenon was observed, and even much sooner after the preparation of solution: the time after which the fractions begin to appear is in case of plasmatic preparations a matter of years—in case of placental preparations of the order of months.

Plasmatic and placental products stored one year at + 4° C. were compared by quantitative precipita-

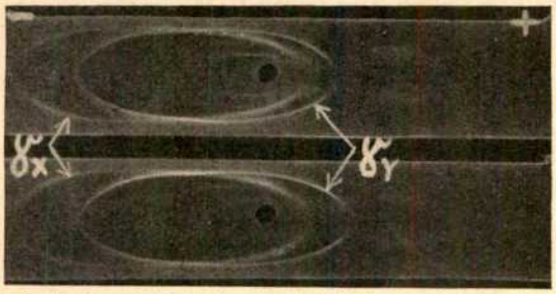

Fig. 1. Immunoelectrophoresis of γ-globulin prepared according to Cohn, aged for 3 yr. at + 4° C.

tion with antihuman antiserum by the capillary tube technique with the following results: plasmatic γ-globulins not yet split were precipitated in a considerably higher dilution (1 : 2,000–4,000) than placental preparations—already split (1 : 32–128). This fact indicates a partial destruction of groups determining the species specificity of the protein in the course of splitting.

The changes in γ-globulins during storage result from a slow proteolytic process. Fractions having identical electrophoretic mobility were obtained by trypsin-, plasmin-, and papain-digestion of a standard γ-globulin. No such manifestations were observed when pepsin-digestion was applied.

The γ-globulin fractions resulting either from storage or from tryptic-digestion can be partially separated by ammonium sulphate precipitation. In Fig. 2 are (from above downwards) immunoelectropherograms of the original outdated product prepared from hæmolysed materials, and fractions appearing after 25 per cent, 25–30 per cent, 30–35 per cent, and 35–50 per cent saturation. At more than 50 per cent only the γ_X component is precipitated. The fraction between 35 and 50 per cent contains a mixture of γ_X and γ_Y component without the original γ-globulin. It is interesting that these two proteins behave as immunologically different antigens, judging from the site of arc-contact according to Ouchterlony's principle[2].

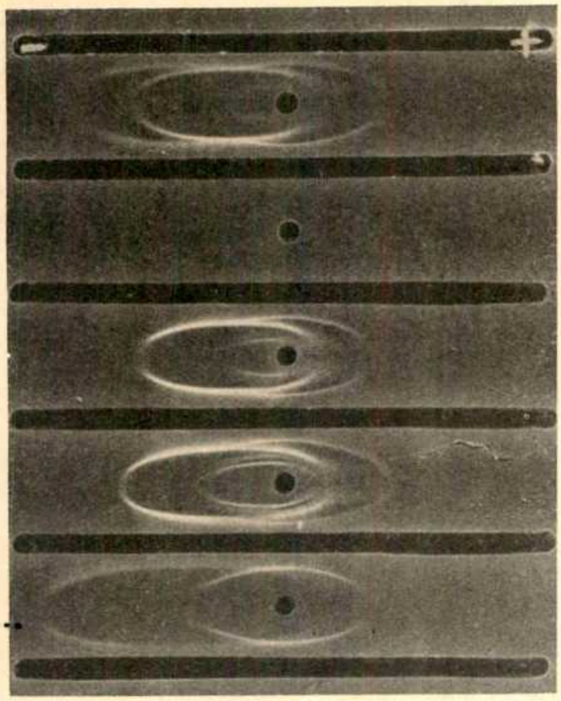

Fig. 2. Ammonium sulphate fractionation of spontaneously split γ-globulin from hæmolysed material

Among the products studied was also a sample of γ-globulin prepared experimentally by Lever's modification of Cohn's method 10 from placentæ[3]. This preparation contained in the final 16 per cent solution approximately 0·3 per cent of sodium tartarate as a remainder from the production technique. In this product both the above-mentioned fractions appeared in the course of a few months accompanied simultaneously by the formation of a considerably large quantity of small needle-like crystals showing a pearly glossy colouring after shaking (Fig. 3). Immunoelectrophoresis of isolated, washed and recrystallized particles revealed that their protein component was formed by the atypical γ_Y fraction. The structure of the crystals and the role of the other ingredients present in the solution will be studied in detail in the future. By ultracentrifugation of the original, spontaneously storage-split product two fractions were isolated, one having S_{20} 4·8, the other 8·2; the solution of the crystalline fraction was homogeneous in the ultracentrifugation, having S_{20} 4·4.

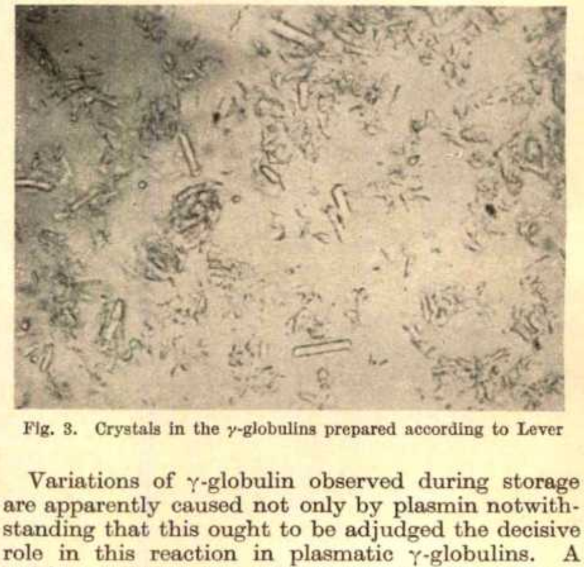

Fig. 3. Crystals in the γ-globulins prepared according to Lever

Variations of γ-globulin observed during storage are apparently caused not only by plasmin notwithstanding that this ought to be adjudged the decisive role in this reaction in plasmatic γ-globulins. A quicker splitting in products prepared from hæmolysed materials indicates the share of 'neutral' enzymes present in erythrocytes[4]. The appearance of crystalline atypical fraction seems to indicate the analogy with γ-globulin fraction III isolated by Porter[5] by papain-digestion of rabbit γ-globulin, but regardless of the crystalline structure.

Further details of our observations will be published elsewhere.

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[1] Gerlough, T. D., Twelfth Conference on Blood Cells and Plasma Proteins, Albany, N.Y. (November 1957).

[2] Ouchterlony, O., *Acta Pathol. microbiol. Scand.*, **25**, 186 (1948); **32**, 231 (1953).

[3] Lever, F. W., Gurd, F. R. N., Uroma, E., Brown, R. K., Barnes, B. A., Schmid, K., and Schultz, E. L., *J. Clin. Invest.*, **30**, 99 (1951).

[4] Morrison, W. L., and Neurath, H., *J. Biol. Chem.*, **200**, 39 (1953).

[5] Porter, R. R., *Nature*, **182**, 670 (1958).

Effect of Amethopterin and 6-Mercaptopurine on the Pattern of Oxygen Tension Measurements in the Fluid Phase of Serum Agar Cultures of Bone Marrow Flecks

We have recently described an electro-chemical method of measuring the oxygen tension in the fluid phase of serum agar cultures of bone marrow flecks[1]. Oxygen tension decreased, at first fairly quickly and then more slowly over a period of three days. This was ascribed to the utilization of oxygen by the surviving cells. In this communication we describe some preliminary experiments upon the effect of amethopterin and 6-mercaptopurine on the oxygen tension pattern of similar cultures. At the beginning of the experiments the culture fluid was equilibrated with air containing 5 per cent carbon dioxide; the carbon dioxide was required to keep the *p*H of the fluid in the physiological range. After a number of hours the fluid was replaced either with fresh medium alone or with medium containing the drugs. As this fluid was also equilibrated with the air–carbon dioxide mixture, the oxygen tension rose but not usually to the starting level. This may have been due to the mixing of the fresh medium with the relatively anoxic serum agar also present in the culture chamber. Thereafter, in the normal controls the oxygen tension continued to fall until the experiment was concluded. This is illustrated by the control curve seen in Fig. 1.

When a medium containing a 1 m*M* concentration of amethopterin was added to the culture of normal marrow there was a definite alteration in the pattern which is illustrated in Fig. 1. After a latent period of about 8 hr. the curve flattened; this we think is due to interference with the normal oxygen metabolism of the cells. It did not occur in 0·25 m*M* concentrations; 0·5 m*M* concentrations showed an intermediate effect.

The findings with 6-mercaptopurine were comparable. We found that a 6-m*M* concentration caused a flattening in the oxygen tension curve after a latent period of about 20 hr.; this did not occur with 2-m*M* concentrations and 4-m*M* concentrations had an intermediate effect.

The oxygen tension patterns of the fluid phase of serum agar cultures of marrow flecks from a case of acute leukæmia have also been studied. The patient, a woman aged forty, was admitted to hospital with pneumonia. The peripheral blood showed a subleukæmic picture, and the bone marrow was stuffed with primitive leukæmic cells. After a stormy course and treatment with antibiotics, steroids and 6-mercaptopurine a remission was induced. This lasted 5 months, after which relapse and death occurred. At no time was the patient given antifolic drugs. At the time of the cell culture the marrow showed predominantly primitive leukæmic cells. Four cultures were made without the addition of drugs, and the oxygen tension patterns resembled those found in normal marrow. 6-mercaptopurine was added to three cultures in concentrations of 6, 4 and 2 m*M*, respectively. The curve obtained with the culture containing the 6-m*M* concentration showed a levelling after about 10 hr.; those obtained from the 4- and 2-m*M* concentrations continued downwards. Amethopterin was added to three cultures in concentrations of 1, 0·5 and 0·25 m*M* respectively; in none of these was a levelling effect seen.

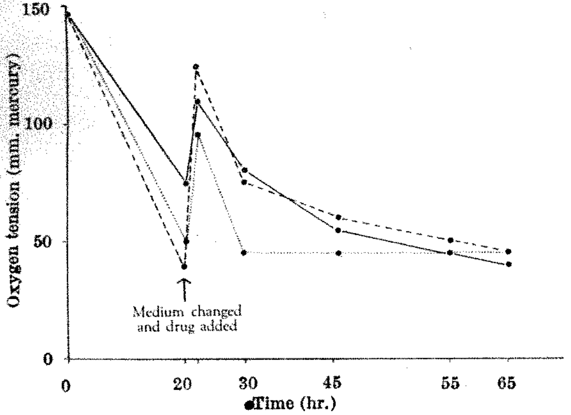

Fig. 1 ●—●, No drug control; ●···●, 1 m*M* amethopterin; ●- - -●, 0·25 m*M* amethopterin

These preliminary results are of interest because they could form the basis of an *in vitro* test of the sensitivity of leukæmic marrow cells to various chemotherapeutic agents. This would be based on the hypothesis that the concentrations of drugs which would inhibit the oxygen uptake of sensitive cells might not affect cells which were resistant.

We are grateful to Messrs. Lederle and Burroughs Wellcome for a supply of drugs.

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[1] Davis, J. M. G., and Woodliff, H. J., *Blood* (in the press).

PLANT PHYSIOLOGY

Effect of Gibberellic Acid on Flower Differentiation in Petkus Winter Rye

It is known that application of gibberellic acid causes flowering of many biennials (*Hyoscyamus*, *Apium*, *Brassica*, *Daucus*, etc.)[1]. In these cases gibberellic acid substitutes for cold (vernalization) treatment. Treatment of winter cereals (Petkus rye[2], barley[3], winter wheat[4]) with gibberellic acid, on the contrary, has been found to have no effect on the induction of normal flowers. Chaylakhian[5] has reported that treatment of non-vernalized oats, variety Victory, with gibberellic acid, under short-day conditions, caused formation of an increased number of developed panicles. This was not the case under long-day conditions. Paleg and Aspinall[3] have reported that treatment of barley with gibberellic acid for several weeks immediately following emergence results in the formation of abnormal spikes.

The experiments reported here are concerned with the normal flower formation of Petkus winter rye by application of gibberellic acid. This cereal is, under the growing conditions of our experiments, a cold-requiring plant.

It has been found that, to be successful, treatment must occur at a particular stage in the development of the plant.

All the experiments were performed under the controlled greenhouse conditions of the Earhart Plant Research Laboratory. A description of this laboratory and the experimental facilities available in it has been given by Went[6].

Plants of winter Petkus rye, grown under a constant temperature of 17° C., both in short-day (8 hr.) and long-day (16 hr.) regimes, were sprayed with water solutions of gibberellic acid (200 p.p.m.). The plants of each day-length regimen were divided into three groups: (1) controls, sprayed with de-ionized water; (2) spray treatment started when the plants had five visible leaves; (3) spray treatment started when the plants had ten visible leaves. Each group was composed of 18 plants. The plants were sprayed for seven successive days after which they were permitted to develop without further treatment.

75 days after sowing the seed the plants were dissected and the stage of development of the growing point of each main shoot recorded. For the five-leaf stage this dissection occurred 48 days after the end of treatment; for the ten-leaf stage, 12 days after treatment. In order to evaluate the stage of development, an arbitrary numerical scale from 0 to 4 was used, in which stage 0 is the early vegetative apex and 4 is a complete flower just prior to full development of the awns.

The results (Table 1) show that winter Petkus rye which has not been vernalized can be induced to form normal flowers by the application of gibberellic acid. The application of the hormone apparently must, however, occur at a proper time in the development of the plant (ten-leaf stage). Applied before this time, gibberellic acid does not induce flowering. It is, on the contrary, inhibitory. The typical effect of gibberellic acid on growth in grasses is found in both the early and the later applications of the acid, that is, elongation of the internodes and narrowing of the leaves.

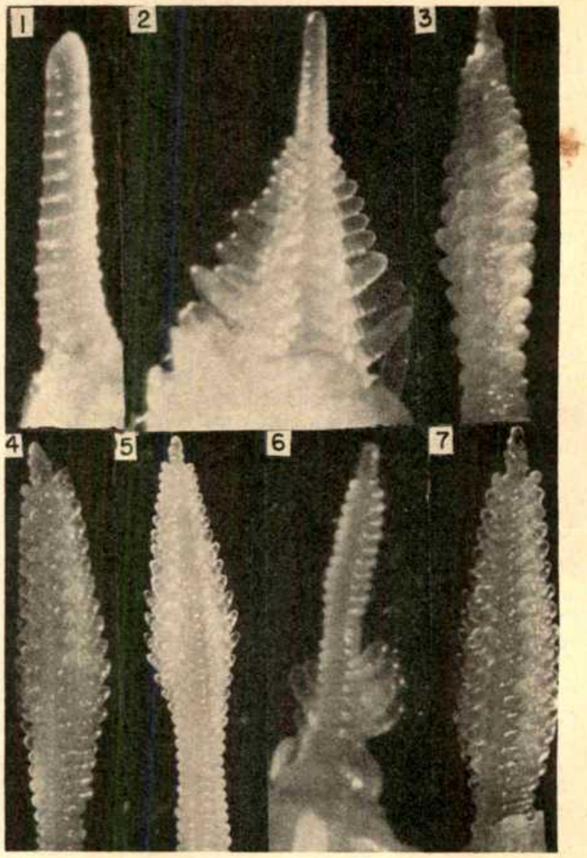
Table 1. EFFECT OF GIBBERELLIC ACID ON THE FLOWERING BEHAVIOUR OF WINTER PETKUS RYE

Treatment with 200 p.p.m. at the	Long day	Short day
Five-leaf stage	Veg./abnormal* 18 plants	Veg./abnormal 13 plants* Flower/abnormal 4 plants 17 plants
Ten-leaf stage	Flower primordia (stages 3–4)* 18 plants	Flower primordia (stage 4 to head)* 18 plants
Control (water spray)	Vegetative* 17 plants	Flower primordia (stages 3–4) 10 plants Veg. primordia 8 plants 18 plants

* See Figs. 1–7 for a description of these developmental stages.

Figs. 1, 2 and 3 are photographs of typical apices of the three treatments on long day. Fig. 1 shows the apex of a control plant after 75 days of growth. The plants are all vegetative. Fig. 2 shows the effect on the growing point of applications of gibberellic acid at the five-leaf stage. It can be seen that the lower part of the apex is abnormal with great numbers of developing tillers. The upper part is normally vegetative. We consider this portion of the apex as having been developed after the effect of the applied gibberellic acid has ceased. Fig. 3 shows a typical apex from a plant treated after the tenth leaf has unfolded. This is a normal flowering apex as can be seen by comparison with Fig. 4, which represents an apex dissected from a vernalized control plant at the same stage of development as that shown in Fig. 3.

Figs. 5, 6 and 7 represent apices dissected from plants grown on short days. Fig. 5 pictures the unvernalized apex of a control plant; because of the promotive effect of short days on flower formation in unvernalized winter rye (short-day vernalization: see ref. 7), this apex is further advanced than comparable apices of plants grown on a long-day regimen and shows an early stage of flower development. Fig. 6 shows the effect of gibberellic acid treatment at the fifth-leaf stage. These plants show a distinct abnormality at the base of the apex with an early stage of a normal flower at the top of the apex. Fig. 7 shows the effect of gibberellic acid treatment at a later stage (10 leaves).



Figs. 1–7. Stem apices of control, vernalized and gibberellic acid-treated plants

On short day there is a significant difference between controls and those plants treated at the ten-leaf stage in the number of plants which flowered at the time of dissection, as well as the stage of development. Only 5 per cent of the controls had well-developed flower primordia, while all the plants treated at the ten-leaf stage had advanced flower primordia. Treatment at the five-leaf stage on short days resulted not only in abnormalities, but also the upper part of the apex, on which flowering had started, was at a significantly earlier stage in development of the flower.

Considering the long-day experiments, it is obvious that the plants must reach a specific stage of development before they can be induced to flower. Purvis and Gregory[7,8] have shown that there is a minimal leaf number (7) before flowering in winter rye can occur. The primordia above the eighth leaf are indeterminate and can produce either leaf or spikelet. In our experiments the plants in which gibberellic acid caused flowering had 10 exposed leaves, which means that the number of leaves and primordia totalled between 20 and 25. Gibberellic acid applied at this stage affects the development of the labile primordia so that flowers rather than leaves are formed. Since gibberellic acid affects flower formation in plants only after the ten-leaf stage, the acid or a similar substance must be limiting, while 'florigen' is not.

In our experiments and especially in those of Paleg and Aspinall[3], it has been shown that applications of gibberellic acid at an early stage in the development (before the ten-leaf stage) will result in inhibition of flowering. Therefore this early application must either inhibit the formation of, or destroy, the florigen.

Hence, at least two things must happen during vernalization: (1) 'florigen' must be formed, or its formation induced after removal from cold; (2) a gibberellic acid-like substance must also be made, accumulated, or its formation induced so that it is formed soon after the plant is removed from the cold.

A more detailed report of this work is being prepared for publication elsewhere.

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[1] Brian, P. W., *Biol. Rev.*, **34** (1), 37 (1959).

[2] Lang, A., *Proc. U.S. Nat. Acad. Sci.*, **43**, 709 (1957).

[3] Paleg, L., and Aspinall, D., *Nature*, **181**, 1743 (1958).

[4] Weibel, D. E., and Futrell, M. C., *Plant Physiol.*, **33**, Supp., 42 (1958).

[5] Chaylakhian, M. K., *Fiziol. Rast.*, **5** (6), 547 (1958).

[6] Went, F. W., "Experimental Control of Plant Growth" (Chronica Botanica Co., Waltham, Mass., 1957).

[7] Purvis, O. N., and Gregory, F. G., *Ann. Bot.*, N.S., **1**, 569 (1937).

[8] Purvis, O. N., *Ann. Bot.*, N.S., **48**, 919 (1934).

Effect of Gibberellin on the Flower Initiation and Stem Extension in Petkus Winter Rye

THE cold requirement for flower formation in biennial *Hyoscyamus niger* was shown by Lang[1] to be replaceable by application of gibberellin, but no flowering response was observed at that time in either Petkus winter or spring rye similarly treated[2]. In 1958, I attempted to supplement partial vernalization in Petkus winter rye by a single injection of gibberellic acid (10 p.p.m.) through the third leaf. No effect on flower initiation was observed. Stem extension was unaffected in the unvernalized controls but considerably enhanced in the partially vernalized series. In 1959, further work was carried out to determine whether the lack of response was due to the low concentration of the single application of gibberellic acid, or whether stimulation of stem extension demanded some prior degree of flower initiation.

Unvernalized winter rye was injected with gibberellic acid at 20 p.p.m.; this was done by feeding through the severed tips of mature leaves. Three groups received single injections to the third, sixth and ninth leaves, respectively, at the time when these had reached full expansion (that is, approximately at intervals of 3 weeks). In a fourth group the sixth and ninth leaves were injected as they reached maturity, while in a fifth group the third, sixth and ninth leaves were treated in turn.

The results are presented in Table 1, in which the effects on progress of flower initiation as 'scores' attained 93 days after planting, and on stem extension, are shown. 'Scores' less than 21 indicate that flower initiation (double ridges) had not been attained, while higher values mark progress beyond this stage.

Stem extension. A single injection at the third-leaf stage had no effect, as was also found in 1958. Single injections at the sixth or ninth leaf-stages resulted in limited stem extension (3 cm. in each case), apparently attained at a higher rate when the ninth leaf was treated. Multiple injections produced an effect rather more than proportional to the number of leaves injected. Thus an injection of the third leaf, apparently ineffective when this leaf alone was treated, intensified the response to treatment of later leaves.

Flowering response. The flowering response was entirely different. Injection of the third leaf resulted in a very small increase in 'score' just below the significant level. Injection of the sixth leaf resulted in a significant increase, which was further augmented by injecting the ninth leaf. Multiple injections effected no greater increase than that following a single injection of the ninth leaf; indeed, the triple injection decreased the score significantly below that due to the single injection.

Although the plants were not grown to maturity, it was evident that those injected through the ninth leaf alone would have produced ears free from morphological abnormalities. Where earlier leaves had been injected in addition, very many abnormalities were found, such as the formation of subsidiary ears from basal primordia ('miracle' ears), leafy outgrowths below the spikelets, or shortening and distortion of the peduncle[3]. This was also true to a lesser degree where the sixth leaf had been injected, but was not, of course, apparent in the undifferentiated apices of plants treated at the third leaf. These effects correspond closely with those obtained by Caso *et al.* (preceding communication).

Thus it appears that gibberellin has no direct effect on flower induction in Petkus winter rye, and therefore cannot replace low temperature in this respect; the accelerating effect on flowering would appear to operate at some post-inductive stage. The stimulus to stem extension only occurs either just before or after flower initiation has taken place.

Table 1. EFFECT OF INJECTING UNVERNALIZED WINTER RYE WITH GIBBERELLIC ACID (20 P.P.M.) THROUGH DIFFERENT LEAVES

No. of injections Leaf injected	0 None	1 3	1 6	1 9	2 6 + 9	3 3 + 6 + 9
Score at time of injection		7·1 ± 0·34	9·6 ± 1·19	13·0 ± 0·91		
		←*NS*→	←*SS*→	←*SS*→	←*NS*→	←*S*→
Score at 93 days	17·8 ± 0·39	19·2 ± 0·78	23·4 ± 0·66	27·8 ± 0·49	27·8 ± 0·40	26·3 ± 0·58
Stem length at 93 days (cm.)	0	<0·5	3·0 ± 0·89	3·1 ± 0·50	7·7 ± 1·22	13·1 ± 1·80

N.S., Difference not statistically significant; *S.*, significant, $P = 0·05$; *S.S.*, highly significant, $P = 0·01$.

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[1] Lang, A., *Naturwiss.*, **43**, 284 (1956).

[2] Lang, A., *Proc. U.S. Nat. Acad. Sci.*, **43**, 709 (1957).

[3] Gott, M. B., Gregory, F. G., and Purvis, O. N., *Ann. Bot.*, N.S., **19**, 87 (1955).

BIOLOGY

Rare-Earth Fatty Liver

THE biological behaviour of the rare earths has recently become a question of increasing importance[1]. Early recognition of the prominent occurrence of several of these elements in fission products[2] stimulated a wide scope of distribution studies in animals and, to a limited extent, in man. Yet, we could find among these studies no mention of a specific biochemical or histological effect except for a brief communication from this laboratory on fatty liver degeneration[3] and some earlier reports on rare earths as anticoagulants[4].

We recently characterized the occurrence of an acute fatty infiltration of liver caused by the stable rare earths of lower atomic number (lanthanum to samarium) when they are administered intravenously to rats. The fatty response, which is represented by an increase in the neutral esterified fatty acids with little change in the phospholipid or cholesterol content, is influenced by a variety of hormonal factors[5]. This communication gives results that show the effect of time on recovery of the rat liver from the lipid disorder and the liver lipid response of certain other species when a rare earth is administered.

Female rats (Carworth Farm-Nelson) weighing 180–240 gm. were used in the time study which is summarized in Fig. 1. Pertinent information for the various species used in the other study is included in Table 1 along with the results. All animals in both experiments were fasted 24 hr. before they were killed. The materials and the methods of extraction and measurement of total liver lipids were the same as those used previously[5].

Fig. 1 shows that the maximal increase in liver lipids after a 2 mgm./kgm. dose of stable cerium occurs at 2–3 days. Six days after receiving the cerium injection the liver lipids returned to normal. The recovery period, the fourth to fifth day, shows a wide biological variation. Some preliminary studies indicate that the amount of cerium in the liver remains at essentially the same high level (about 70 per cent of the administered dose) at least from 4 hr. to 12 days (Rafter, J. J., Kyker, G. C., and Snyder, F., unpublished results); that is, there is no tendency for the liver cerium concentration to decrease during the recovery phase.

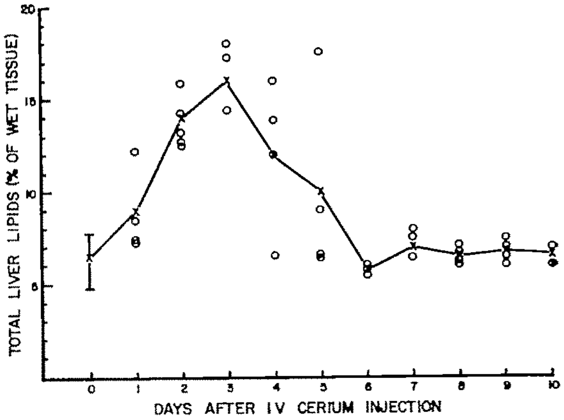

Fig. 1. Total liver lipids as a function of time after intravenous cerium (2 mgm./kgm. in rats). ○, Individual rat value; ○, two individual rat values are identical; ×, mean value. The range is shown for the control group (0 days) with the mean value based on analyses of 18 rats

Because cerium appears to be rather representative for studying the behaviour of the first part of the rare-earth series, this element has been used to survey the lipid response in both males and females of other species under the same conditions that had caused the fat disturbance in rats. The results in Table 1 show a significant elevation in liver lipids for rats (at 3·5 mgm./kgm. for females and 14·0 mgm./kgm. for males), mice (at 7·0 and 14·0 mgm./kgm. for females), and hamsters (at 7·0 and 14·0 mgm./kgm. for males). No fatty livers were seen in any of the other species except in two male rabbits that had 14·7 and 18·2 per cent liver lipids at 48 hr. after a 3·5 mgm. cerium dose. This variation in

Table 1. LIVER LIPIDS IN DIFFERENT SPECIES 48 HR. AFTER A SINGLE INTRAVENOUS CERIUM INJECTION

Species	Sex	Cerium (mgm./kgm.)				
		0	3·5	7 (per cent lipids in wet tissue*)	14	28
Rat (200–267 gm.)	M	5 34 (3) 5·08–5·69	7·93 (3) 6·47–10·74	9·54 (3) 8·85–10 29	9·16† (3) 8·33–10·33	
	F	6·11 (7) 4·88–7·40	11·35† (12) 9·39–14·53			
Mouse (13–20 gm.)	M	7·09 (2) 7·02–7·16	6·99 (3) 5·57–9 62			
	F	6·01 (6) 4·67–7·42	9·01 (7) 5·51–12·67	17·52† (4) 9·47–24·53	17·00† (4) 15·49–20·73	
Hamster (80–127 gm.)	M	6·39 (7) 5·34–7·25	6·91 (4) 4·70–8·04	12·85† (5) 7·26–16·15	8·90† (4) 7·06–14·00	6·22 (7) 5·85–7·04
	F	6·22 (3) 6·08–6·48	5·09 (4) 4·76–5 58	5·59 (3) 5·34–6 08	7·27 (4) 4·97–9 90	5·14 (3) 4·55–5·60
Rabbit (1,910–2,420 gm.)	M	6·38 (5) 5 02–9 36	9·39 (6) 5·41–18 24	7·51 (3) 5·00–9 34	6·50 (3) 5·69–7·51	
	F	5·44 (6) 4·39–6·98	7·34 (7) 5·35–11·71	5·25 (3) 4·48–6 50	4·87 (3) 4·68–5·03	
Guinea pig (315–708 gm.)	M	8·09 (9) 5·42–10·89	7·05 (4) 6·10–7·87	7·68 (5) 5·40–10·35	7·13 (4) 6·33–7·50	7·82 (5) 7·68–9·57
	F	7·40 (9) 4·71–9·76	6·49 (4) 4·80–8·25	8·77 (5) 5·47–12·69	8·23 (5) 6·89–9 56	7·71 (6) 6·79–9·12
Chicken (1,230–1,770 gm.)	F	5·06 (8) 4·69–5·40	5·16 (6) 4·57–7·10	4·94 (5) 4·60–5·31	5 03 (4) 4·86–5·28	
Dog (8,000–12,000 gm.)	F	5 46 (1)	4·52 (1)		7·70 (1)	4·00 (1)

* The tabulated values represent consecutively the following: (1) the average of analytical results on individual animals in a group; (2) the number of animals in a group (shown in parentheses); (3) the range of observations on individual animals (hyphenated).

† In the interpretation of the difference between the mean of the control group (0 mgm./kgm.) and the cerium-treated groups (3·5–28 mgm./kgm.) of that species, the test of significance was applied to all the results. The standard deviations and the P values are not tabulated along with the results because of the small number of animals in the groups (220 animals; 48 groups). The dagger does signify for six groups (rats, mice and hamsters) a highly significant difference ($P < 0·01$) when interpreted by usual procedure in statistical analysis.

response among the seven species that were studied may be due either to biochemical species differences or to differences in time of response.

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[1] Kyker, G. C., and Anderson, E. B. (ed.), "Rare Earths in Biochemical and Medical Research", U.S.A.E.C. Rep., *ORINS*-12 (1956).

[2] Hunter, H. F., and Ballou, N. E., *Nucleonics*, 9 (5), *C*2–*C*7 (1951).

[3] Kyker, G. C., Cress, E. A., Sivaramakrishnan, V. M., Steffee, C. H., and Stewart, M., *Fed. Proc.*, 16, 207 (1957).

[4] Steidle, H., "Seltene Erdmetalle", in Heffter, A., and Heubner, W., "Handbuch d. experiment. Pharmakologie", 3 (Pt. 4), 2189 (1935).

[5] Snyder, F., Cress, E. A., and Kyker, G. C., *J. Lipid Res.*, 1, 125 (1959).

Quantitative Estimations of Populations of the River Crab, *Potamon* (*Potamonautes*) *perlatus* (M. Edw.), in Rhodesian Trout Streams

THE river crab is the largest invertebrate to occur in Rhodesian trout streams, but standard quantitative sampling methods reveal insignificant numbers of this animal. As has been suggested by Van Someren in Kenya[1], it is doubtful if anglers have been correct in assuming that the crab is an important trout-food item. Nevertheless, preliminary Rhodesian studies[2] indicate that it occurs in trout stomachs with a median percentage frequency occurrence of 13·1; furthermore, there is a 100·0 per cent frequency and 97·5 per cent volumetric occurrence in droppings of the clawless otter, *Aonyx capensis* Schinz. These figures are based upon slightly more than 1,000 stomach contents, and exactly 1,000 otter samples. In such circumstances the river crab takes on a certain economic standing, even if ignored as a competitor or scavenger.

The known Rhodesian habitat of *P. perlatus* (var. *inflatus*, *depressus*, *sidneyi*) can be described as mountain streams having an altitude ranging from 5,000 to some 6,800 ft. a.m.s.l. (1,520–2,070 m.), although the species may well be found both above and below these limits in addition to occurring in weirs or dams built upon the streams. In these waters the crabs can be taken in funnel-entrance traps baited with fresh fish, and if such traps are enclosed within a screened but typical section of stream then the number of animals captured decreases each time the traps are cleared. Thus it is possible to apply the population estimation method of De Lury[3], in which numbers of animals captured are plotted graphically against an ordinate, and total catch to date against an abscissa. Extrapolation of a fitted straight line to an intersection with the abscissa provides an estimation of the population present at that time. In practice, traps are cleared each 24 hr., and there is little difficulty in relating the results of test areas to the total area of stream-bed under consideration.

Chemical destruction of soft and hard parts of adult crabs suggests that the wet weight of soft parts represents but about 34·0 per cent of total wet weight. In assessing populations as potential fish or otter food this factor should be taken into account. It may be recalled that Allen[4] treated the mollusc *Potamopyrgus* in similar fashion, although that worker dealt with dry weights.

Trapping is selective in that small specimens having a maximum carapace width of less than 17·0 mm. are not attracted to baits so far tested. However, the larger size groups consistently taken in traps can usefully be discarded when taken in orthodox net or dredge samples. In impoverished African trout waters the occurrence of a single large crab can have a grossly distorting effect upon the collection, and when such large-size animals appear it is an asset to be able to ignore their occurrence. The small-size group, in Rhodesian trout streams being less than 17·0 mm. carapace width, have less distorting effect and may be incorporated in orthodox sampling results. During practical field work it is therefore suggested that trapping forms a valuable adjunct to a series of dredge and net samples, provided that the degree of selectivity is determined and allowed for.

Preliminary results at Inyanga, the main trout area of Central Africa and having a central point at about 32° 44′ E., 18° 17′ S., indicate that *perlatus* specimens subject to trapping range from 17·3 to 45·0 mm. maximum carapace width. The range for males is 22·0–45·0 mm., and that for females 17·3–37·5 mm. The median wet weight of all specimens taken, after drying-off of all surplus moisture, ranges from 7·375 to 10·510 gm. There is some evidence that smaller size groups pertain where other trout food organisms are in poor supply; but in these areas the crabs are infested with a small greenish leech, normally some 10·0 mm. in length when extended, and customarily found at the bases of the legs. To date no phoresic associations with the crabs have been detected, such as the phoresis between *Potamon niloticus* and immature stages of *Simulium neavei*, and between a crab species and *S. nyasalandicum*[5]. Calculated populations of river crabs in Rhodesian trout streams range from 3,049 to 5,227 per acre (7,530–12,910 per hectare). The wet weight per acre ranges from 49·5 to 121·2 lb. (55·5–135·8 kgm. per hectare), and the wet flesh weight from approximately 16·82 to 41·2 lb. per acre (about 18·86–46·15 kgm. per hectare).

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[1] Van Someren, V. D., "The Biology of Trout in Kenya Colony" (Govt. Printer, Nairobi, 1952).

[2] Turnbull-Kemp, P. St. J., "A Preliminary Survey of the Trout Fisheries of the Federal National Parks" (Rep. to Director of Federal National Parks, Salisbury, 1956).

[3] De Lury, D. B., *Biometrics*, 3, 4 (1947).

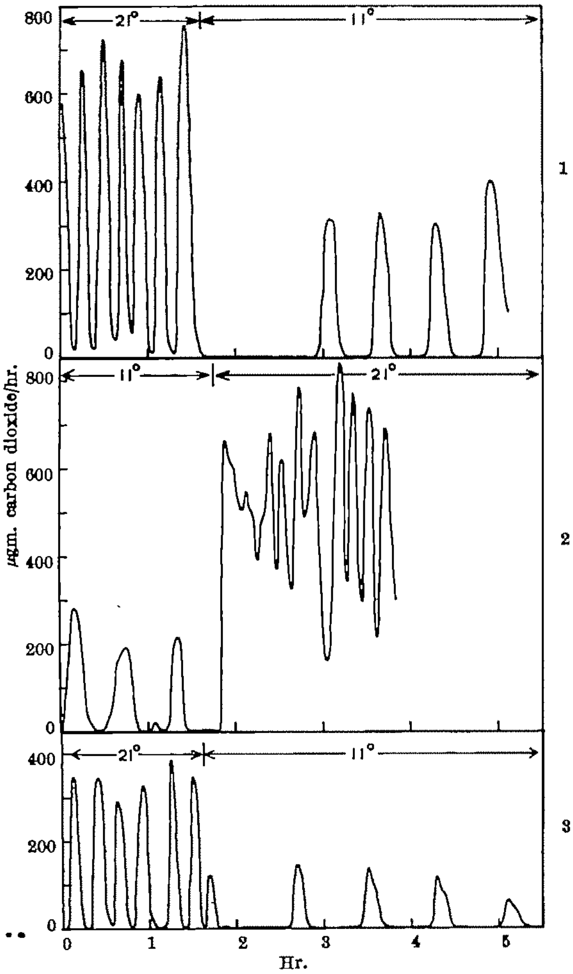
[4] Allen, K. Radway, N.Z. Marine Dept., Fish. Bull. No. 10 (Wellington, New Zealand, 1951).

[5] Freeman, P., and de Meillon, B., "Simuliidae of the Ethiopian Region" (British Museum (Natural History), 1953).

A Temperature-dependent Endogenous Rhythm in the Rate of Carbon Dioxide Output of *Periplaneta americana*

Ball, Dyke and Wilkins[1] and Wilkins[2] developed an apparatus, incorporating an infra-red gas analyser (Grubb-Parsons, *S.B.*1), for continuously estimating the rate of carbon dioxide metabolism of plant tissue maintained under constant external conditions for 5–10 days. Using this apparatus the rate of carbon dioxide output of individual adult cockroaches was determined. The insects were kept in 12 : 12 hr. alternations of light and darkness and, at the commencement of an experiment, were transferred to a small flask which was placed in darkness and at a constant temperature at the end of a light period. The records presented here were derived from one insect, but five other cockroaches gave closely similar results.

In a stream of air initially free from carbon dioxide (relative humidity 80–90 per cent), darkness and at 21 ± 0·1° C. the period of the rhythm was 14·1 ± 0·8 min. (Fig. 1). On lowering the temperature to 11 ± 0·1° C. carbon dioxide output ceased for 76 min. and then the rhythm recommenced with a period of 35·8 ± 1·4 min. and with an amplitude approximately half that exhibited at 21° C. In an initially carbon dioxide-free air stream the period shows a $Q_{10} = 2·5$. The reverse procedure, increasing the temperature from 11° C. to 21° C., reduced the period from 35·8 ± 1·4 min. to 12·6 ± 1·9 min. (Fig. 2). A lag of 30 min. occurred between the change in temperature and the resumption of the rhythm, and the peaks during the first hour at the higher temperature were somewhat irregular. This may account for the slightly shorter period and larger standard error.



Figs. 1–3. The effect of temperature variation on the rhythm of carbon dioxide output of an adult cockroach maintained in darkness and in either a carbon dioxide-free (1 and 2) or normal (3) air stream. For explanation, see text

In a stream of normal air, darkness and at 21° C. the period of the rhythm was 16·2 ± 2·0 min. (Fig. 3). On reducing the temperature to 11° C. carbon dioxide emission ceased for 45 min. and afterwards recommenced with a period of 47·3 ± 0·03 min. In normal air the period shows a $Q_{10} =$ 2·9 and again the amplitude was halved by the temperature reduction.

Removal of carbon dioxide from the air stream has no significant effect upon the period of the rhythm at 21° C., but at 11° C. the period is significantly longer in a normal air stream than in a carbon dioxide-free air stream. The rhythm was not inhibited or the period modified when insects maintained in an initially carbon dioxide-free air stream were illuminated with white light at an intensity of 3,000 lux.

These results contrast with those of Punt[3], who showed that *P. americana* lacks a rhythm of carbon dioxide emission, output being irregularly continuous in darkness, at 15° C. On the other hand, a rhythm of carbon dioxide output has been reported in the locust[4] in which the period was 2·5 min. in darkness, at 32° C. and in an initially carbon dioxide-free air stream. The locust exhibited the rhythm for only 24 hr. after shedding the last hopper skin. In contrast, the rhythm persisted for at least 4 days in the adult cockroaches used in this work. The period of the rhythm of emission of carbon dioxide in silk-worm pupæ has been shown to vary between 45 and 120 min. at 25° C.[5,6].

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[1] Ball, N. G., Dyke, I., and Wilkins, M. B., *J. Exp. Bot.*, 8, 339 (1957).

[2] Wilkins, M. B., Ph.D. thesis, University of London (1958).

[3] Punt, A., *Physiol. Comp. et Oecol.*, 2, 59 (1950).

[4] Hamilton, A. G., *Nature*, 184, 367 (1959).

[5] Schneiderman, H. A., *Nature*, 177, 1169 (1956).

[6] Levy, R. I., and Schneiderman, H. A., *Nature*, 18 491 (1958).

'Watery Stipe' of Cultivated Mushrooms

In a recent paper, Lindberg[1] described a transmissible disease of *Helminthosporium victoriae* in which the infective agent, not identified, caused a stunting of the mycelium. This condition could be transmitted to healthy cultures through contact, either by growing the two types together or by treating normal cultures with a suspension of macerated diseased mycelium in sterile water.

A very similar condition has now been found in the cultivated mushroom, associated with a disorder known to growers as 'watery stipe', 'brown disease' or 'La France disease'. The macroscopic symptoms are variable and include the death of immature sporophores which may turn brown. Different degrees of water-logging of the tissues, particularly of the stipe, also occur. In mild cases this appears as translucent streaks on the stipes, but in extreme cases the whole sporophore may be saturated and exude drops of liquid from the surface of the pileus. Sporophore distortions, such as elongated and bent stipes, are commonly found. Cropping is much below normal and ceases prematurely, this being the most serious aspect of the disorder to the commercial grower. The variable symptoms have made positive macroscopic identification impossible. The only consistent feature which has been discovered has been the degeneration of the mycelium, which is not always apparent from a cursory examination. If, however, fragments of the mycelium are plated out on an agar medium they will, if infected, produce weak, buff-coloured adpressed colonies without rhizomorphs. Healthy mycelium produces fluffy, white, more rapidly growing colonies with rhizomorphs radiating from the point of inoculation. The cells of the abnormal mycelium are shorter than normal and have a slightly swollen appearance. No other differences have been noted.

This mycelial degeneration has been transmitted to healthy cultures using methods similar to Lindberg's. With the double inoculation method, diseased and healthy cultures grow until their mycelia ultimately come into contact, but anastomoses between the hyphæ of the two colonies have not been seen. The coalescence of the two cultures is slow, as if there were antagonism between them, which is in marked contrast to the behaviour of two healthy colonies, where many of the rhizomorphs fuse. The growth of the healthy culture is usually weaker after coming into contact with the abnormal one; but there is no visible change in the appearance of the older mycelium. However, if this (originally healthy) mycelium is subcultured, its growth is abnormal.

Experiments with macerated mycelium have so far been confined to the use of suspensions containing fairly large fragments of mycelium which are capable of regeneration. Normal mycelium dipped in such a suspension grows at less than half the speed of mycelium dipped in suspensions of healthy mycelium or sterile water. Filtration to remove these mycelial fragments also removes the infective agent.

This investigation into the cause of the stunting of mushroom mycelium continues. Tests for the presence of bacteria have given negative results so far. The presence of a virus may be a possibility, although none has been found in fungi.

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[1] Lindberg, G D., *Phytopath.*, 49, 29 (1959).

GENETICS

Self-Incompatibility System in Two Mexican Species of *Solanum*

So far the genetics of self-incompatibility has been described in only one species of *Solanum*, namely, *S. chacoense*[1]. It is an extremely variable species which occurs in north and central Argentine, Paraguay, Uruguay and south Brazil and now includes *S. caldasii*, *S. subtilius* and *S. jujuyense*[2]. *S. chacoense* has a one-gene, multi-allelic, gametophytic system of self-incompatibility similar to that which has been found in three other genera of the Solanaceae, *Nicotiana*, *Petunia* and *Lycopersicon*. Recently I have found that *S. megistacrolobum* has the same system as its sister South American species *S. chacoense* (in the press), whereas the two Mexican species, *S. ehrenbergii* and *S. pinnatisectum*, have a two-gene system.

Progenies of 20 plants were grown from reciprocal crosses between unrelated clones of each species. Siblings within each family were crossed in all directions and each sibling was also crossed reciprocally with its parents. In *S. ehrenbergii* one family produced four intra-sterile groups of plants, but the family from the reciprocal cross produced six such groups. In *S. pinnatisectum* both the families from the reciprocal crosses gave six intra-sterile groups each. Different groups in one family were cross-compatible with some and cross-incompatible with other groups. There were reciprocal differences in the results of crosses between different groups.

The results obtained in the two Mexican species are conspicuous for being quite different from those usually obtained in the *Nicotiana* type system. They are explained on the basis of the following assumption. The genetics of incompatibility is controlled by two independently segregating loci each with a series of multiple alleles. Pollen determination is gametophytic and there may be individual action or epistasis between the alleles of the two series. In the style there is individual action of S alleles. A pollen grain is incompatible on a style when either one or both of its two alleles (of two different series) are also present in that style. This system may prove to be of general occurrence in the Mexican species and limited to them. It has been found to occur also in *Physalis ixocarpa*[3], a native of Mexico and Central America. In contrast, the 1-locus system appears likely to be confined to the species of South America.

The occurrence of the 1- and 2-locus gametophytic systems of incompatibility in the same genus *Solanum* is of considerable evolutionary interest. There is enough evidence to suggest that the 1-locus *Nicotiana* type of gametophytic incompatibility, occurring in a large number of flowering plants, was the first system of incompatibility to develop, and evolved early in the evolution of angiosperms[4]. The sporophytic system[4] and the 2-locus gametophytic system[5] were both derived from this system. It also seems justifiable to assume that the 2-locus system was able to evolve in the Mexican diploid tuberous *Solanums* because of their geographical isolation as a result of the separation of North and South Americas.

Some of the biological forces which might have led to this genetic distinction between the species of the two groups can be suggested. Several investigators[6], on the basis of cytological studies, have concluded that the basic chromosome number in the family Solanaceae is 6 and not 12, which occurs in most of its species. If it is assumed that the present basic number 12 for a large part of this family has been derived by doubling of the original basic number six, very early in the evolution of this family, then the two S loci in the Mexican species are the result of the duplication of the original S gene. However, the possibility of a segmental interchange bringing about duplicate S loci cannot be ruled out.

Whatever might have been the origin of two S loci, in the beginning they must have caused self-compatibility due to the competition interaction between the S alleles in the pollen[7]. The restoration of self-incompatibility could then have occurred by different methods in the two groups of geographically isolated Solanaceous species. (1) In the South American species having a 1-locus system of incompatibility, the restoration of incompatibility occurred by a selection for the loss of function of one of the two S loci—a phenomenon which seems to have occurred in some of the known tetraploid species (for example, *Trifolium repens*[8]) of other families. (2) In the Mexican species having two S loci, the restoration of incompatibility occurred by a selection against competition interaction between the S alleles in the pollen, thus retaining two functional S loci. The absence of competition interaction in the diploid pollen of *Physalis ixocarpa*[8] seems to support this hypothesis. The reason why the Mexican group of *Solanum* species should have evolved differently in this respect than their sister species in South America is, however, not clear.

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[1] Pal, B. P., and Nath, P., *Nature*, 149, 246 (1942).
[2] Hawkes, J. G., Ann. Rep. Scottish Plant Breed. Sta., 38 (1956).
[3] Pandey, K. K., *Amer. J. Bot.*, 44, 879 (1957).
[4] Pandey, K. K., *Evolution* (in the press).
[5] Lundqvist, A., *Hereditas*, 44, 193 (1958).
[6] Swaminathan, M. S., and Howard, H. W., *Bibliogr. genet.*, 16, 1 (1953).
[7] Lewis, D., *Heredity*, 1, 85 (1947).
[8] Pandey, K. K., *Genetics*, 41, 327 (1956).

PSYCHOLOGY

Intelligence, Personality and Brain Rhythms in a Socially Isolated Community

SIGNIFICANT correlations between alpha rhythm in the electroencephalogram and ratings of the temperament variable primary-secondary function have been attributed to genetically determined individual differences in a central nervous excitability characteristic[1,2]. Since both alpha frequency[3] and vocabulary score[4] of the Wechsler Adult Intelligence Scale were reported as positively correlated with speed of perception, it seemed possible that performance on the Wechsler Adult Intelligence Scale might be influenced by primary-secondary function. This argument was supported by the finding in 34 subjects of a significant correlation ($r = 0{\cdot}51$, $P < 0{\cdot}01$) between alpha frequency and total score on the Wechsler Adult Intelligence Scale. Further analysis suggested that the Scale measures non-intellective in addition to intellective aspects of intelligence, and that the former possess a number of electroencephalogram correlates[5]. The fact that previous studies had failed to reveal correlations between alpha rhythm and intelligence was attributed to differences between the Wechsler Adult Intelligence Scale and other intelligence tests, and also to a sampling factor, it being proposed that the emergence of electroencephalogram correlates of non-intellective components of intelligence is dependent on the intellective and socio-economic homogeneity of the sample studied[6].

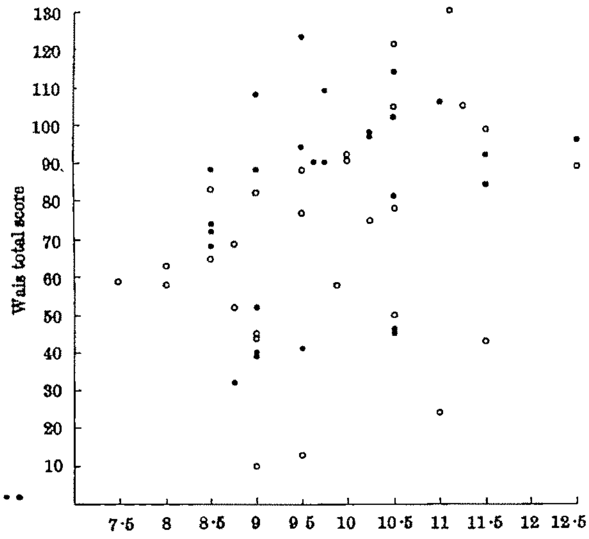

Fig. 1. Correlation chart showing relationship between intelligence score and alpha frequency. ●, Normal electroencephalograms; ○, abnormal electroencephalograms

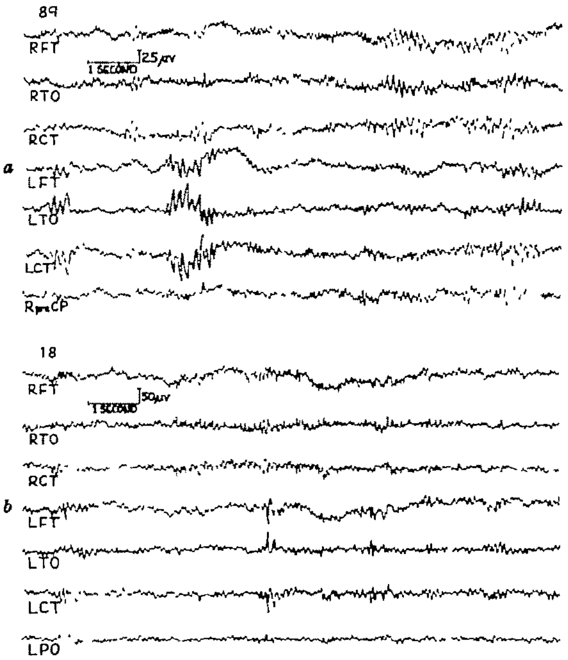

Fig. 2. Electroencephalogram abnormalities from two members of a socially isolated community, neither with any obvious neuropsychiatric symptom or known abnormal history: *a*, male, 64 yr., intelligence quotient not measured. Bilateral independent focal bursts from temporal lobes, more marked on left, frequencies mainly between 8 and 12 c./sec. with some theta rhythm; *b*, male, 56 yr., intelligence quotient 75. Focal spikes and sharp waves from left temporal lobe, also runs at 10–13 c./sec. and traces of theta rhythm. *R*, Right; *L*, left; *F*, frontal; *T*, temporal; *O*, occipital; *C*, central; *P*, parietal; pre*C*, precentral

As a test of these hypotheses we undertook an expedition to a remote region of South Africa to study a group of white labourers from a socially isolated community of long standing, the members of which are characterized by a high degree of behavioural uniformity and generally low intelligence. Electroencephalograms were recorded from 94 men and 2 women, of whom 55 men were also given the South African version of the Wechsler Adult Intelligence Scale. A number of other psychological tests was also applied.

Two important findings have now emerged. First, as predicted, alpha frequency is significantly correlated with total score on the Wechsler Adult Intelligence Scale ($r = 0{\cdot}34$, $P = 0{\cdot}01$, Fig. 1) and with raw scores from 5 Scale sub-tests, including vocabulary. These correlations were virtually unchanged with age held constant. Secondly, 49 (51 per cent) of the 96 subjects possessed abnormal electroencephalograms, all but one because of bilateral shifting foci from the temporal lobes, usually in the form of sharp waves, spikes, or bursts of activity ranging in frequency from 2–14 c./sec. (Fig. 2). These focal disturbances tended to be more prominent from the left side.

The correlations between alpha frequency and Wechsler scores support our hypothesis that given a homogeneous sample it is possible to identify a non-intellective component of intelligence which is related to a central nervous excitability factor. Detailed reports of distributions and interrelations between electroencephalogram variables, Wechsler scores and other psychological measures will be published elsewhere.

The finding of 50 per cent bilateral independent temporal lobe foci was quite unexpected, since this type of electroencephalogram abnormality is strong indication of organic disturbance in one or both temporal lobes, and is particularly common among certain epileptics[4]. Our sample contained one obvious case of epilepsy in the abnormal electroencephalogram group and two possibles in the normal group. There was no relation between any aspect of clinical history as elicited during interview and electroencephalogram abnormality, although no medical examination was performed. Family history revealed relatives with epilepsy in two cases in the normal electroencephalogram group and five in the abnormal group. This difference is not significant, although it seems that the incidence of epilepsy in this community is higher than in the rest of the Union of South Africa. Electroencephalogram abnormality was unrelated to age (Table 1), although the mean age of the abnormal group was significantly lower than the normal group (36 years as opposed to 43).

Table 1. AGE DISTRIBUTION: NORMAL AND ABNORMAL ELECTROENCEPHALOGRAMS

Age group	18–20	21–30	31–40	41–50	51–60	61–70	71–78
Normal electroencephalogram	0	8	8	16	8	4	2
Abnormal electroencephalogram	2	10	12	11	8	6	0

Foremost among possible explanations of the high incidence of electroencephalogram abnormality is birth injury, in view of the inadequacy of obstetric facilities in this region. In addition, malnutrition is common and may be critical, since we found that 36 per cent of a group of African infants with kwashiorkor exhibited temporal lobe foci[7]. Another possibility relates to the effects of inbreeding, known to have been prevalent in this community for many generations. In the words of the local district surgeon, "these people have remained a community within a community, merrily producing misfits of variation". Unfortunately, the need to secure the confidence of our subjects rendered it impossible to obtain any direct information concerning interfamilial marriages, but it was notable that a number of men were married to women with the same maiden name, and among the 91 male subjects showing neither filial nor fraternal relationship the total number of surnames was only 44, of which one occurred 11 times, one 8, three 5, two 4, three 3, and eight twice. This may be compared with 100 members of this Institute, among whom the total number of surnames is 90, of which three are repeated thrice and four twice. These figures support the contention that inbreeding has been prevalent among the population from which our experimental sample was drawn. That the high incidence of electroencephalogram abnormality may in some way be related to a genetic influence cannot therefore be excluded.

The mean intelligence quotient was 75, standard deviation 15, the distribution being skewed towards the lower end of the scale, with 15 mental defectives and 17 borderline mental defectives. However, although the mean intelligence quotient of the abnormal electroencephalogram group was 10 points lower than that of the normal electroencephalogram group, this difference is not significant ($t = 1{\cdot}29$, $P > 0{\cdot}20$). It is therefore unlikely that the low standard of intelligence and the high incidence of electroencephalogram abnormality are directly related consequences of a common factor.

Although distinctive personality attributes and mental symptoms have been commonly reported with electroencephalogram temporal lobe disturbances[6,8], we noted no such differentiating behavioural reactions among our normal and abnormal electroencephalogram groups. However, it should be stressed that the sample is peculiar in that it is drawn from a community which exhibits highly characteristic social traits, for example, an abject satisfaction in things as they are, an absence of individuality and self respect, a blissful self-satisfaction, and a stubborn resistance against any attempt to alter their way of life. It is possible that in such an undifferentiated and socially withdrawn community the development of mental symptoms as often observed elsewhere with temporal lobe foci would be either inhibited or moulded into the cultural framework.

We hope that later we will be able to revisit this region, both to extend the investigation in the neuropsychiatric sphere and to study younger generations, whereby we might evaluate the contribution of heredity to these findings. In the meantime, the observation that 50 per cent of a group which does not complain of fits or obvious neuropsychiatric disorder yet exhibits bilateral shifting foci from the temporal lobes merits careful consideration in relation to neuropsychology.

The expedition was undertaken with our Carnegie Mobile Psychological Laboratory, our colleagues being Miss B. Kirk-Cohen, Miss M. Oertel and Mrs. J. M. Nelson.

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[1] Mundy-Castle, A. C., *J. Nat. Inst. Personnel Res.*, 6, 95 (1956).
[2] Mundy-Castle, A. C., *J. Nat. Inst. Personnel Res.*, Monogr. Supp. 2 (1958).
[3] Mundy-Castle, A. C., *J. Nat. Inst. Personnel Res.*, 6, 38 (1955).
[4] Livson, N., and Krech, D., *J. Pers.*, 25, 46 (1956).
[5] Mundy-Castle, A. C., *J. Pers.*, 26, 184 (1958).
[6] Bingley, T., *Acta Psychiat. et Neurol.*, Supp., 120, 33 (1958).
[7] Nelson, G. K., *EEG Clin. Neurophysiol.*, 11, 73 (1959).
[8] Rey, J. H., Pond, D. A., and Evans, C. C., *Proc. Roy. Soc. Med.*, 42, 17 (1949).

Syllabic 'Strategies' in Written Language

LANGUAGE, whether written or spoken, is one of the most sensitive indices of central nervous disorder; in a variety of neuropsychiatric states the speech and writing of the patient may be the main physical manifestation of disease. A technique of language analysis has been evolved with clinical needs in mind; but an attempt has also been made to provide a neurophysiological working hypothesis to account for the main empirical findings of the method.

The subject is asked to write 'about a page' on any topic of his own choice. The words written are then counted according to their length in syllables (S), and if p_S is the proportion of S-syllable words in the utterance, a quantity termed the 'avoidance' (A_s) is computed for each word-length. Avoidance is defined by the equation: $A_s = -\log p_s$.

The avoidances are then plotted against various simple functions of S. When it is found that A_s is an exact function of S, f(S), over the whole range of word-length employed, the sample is said to obey a pure 'strategy'. Not all, but about 25 per cent, of samples appear to show this phenomenon. Tables 1–4 contain examples of the four pure strategies so far encountered, which have been named and formulated as follows:

Linear: $A_s = k_1.S - A_0$ Logarithmic: $A_s = k'.\log S + A_0$
Quadratic: $A_s = k_2 S^2 - A_0$ Log. factorial: $A_s = k_f \log S! + A_0$

In these equations the k are slope-constants and the A_0 are intercepts on the A_s axis, usually with the signs indicated. A_0 is found to be zero in some utterances; in such cases the strategy requires only one constant to specify it and is therefore termed 'primitive'. In the Tables A_s are observed values while $A_s{}^*$ are values read off from a line drawn by eye through the points. Comparison of the two columns will indicate the closeness of fit of the strategy-equations to the data.

It seems that the brain tends to fall into a relatively limited number of patterns with respect to its spontaneous distribution of word-length. Some samples obey no obvious strategy; others show imperfect fit to one of the pure strategies; others can be described as obeying 'mixed' strategies which are combinations of pairs of the four pure types.

The likelihood that a subject will reproduce the same strategy on two separate occasions is still uncertain; but several subjects who have written two samples, separated by time-intervals ranging from minutes to days, in the absence of any apparent major change in physical or mental state during the interim, have shown stability of strategy apart from variations in the constants k and A_0. However, physicochemical agents which cause acute changes in the clinical state of the brain seem able to bring about 'switches' from one strategy function to another.

The findings can be explained by the following provisional hypothesis: The utterance of language consists in the conversion of specific patterns of neural activity corresponding to conceptual states (thoughts) into other patterns capable of eliciting appropriate motor activity from the motor effectors. This recoding process is assumed to involve several distinct information-handling operations, all of which must be completed before each unit of language can be uttered; hence the slowest operation will be rate-limiting for the overall process of uttering language units. The key postulate of the theory is that if the same operation remains rate-limiting for the whole of an utterance, the character of that operation will determine the strategy.

The assumption is also made that the avoidance (that is, statistical information) of S-syllable words is proportional to the amount of information handled by the neurones performing the operation which is rate-limiting for words of this length.

Now strategies are defined above in such a way that the *word* is the unit of language and syllables are sub-units. To specify a word it is necessary to specify both its component syllables and their order. Suppose that in a given utterance the rate-limiting process (for all word-lengths) happens to be that of ordering the syllables. A set of S syllables can be arranged in factorial S ($S!$) different orders, of which one is correct and must be selected. The information required for a discrimination of this complexity is, ideally, proportional to $\log S!$. On the above assumptions, the avoidance should be given by: $A_S = K.\log S!$. This is the equation of a primitive log factorial strategy.

In a similar way it may be imagined that other operations (such as the formulation of ideas) are required in the generation of an utterance and could become rate-limiting, so giving rise to other strategies. The constants A_0 are not explained by the theory in its simple 'ideal' form but may be due to the imperfection of the brain as a communication mechanism. Mixed strategies and strategy-switches are explained by natural extensions of the theory. A fuller account of this work is in preparation.

The collection of material has been greatly helped by a grant from the Theodore Chapin Beebe Fund. I am indebted to Dr. Hick of the Cambridge Psychological Laboratory and to Dr. Russell Davis of the Medical Psychology Laboratory for their constructive criticism and advice.

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Oct. 7.

Table 1. LINEAR STRATEGY

Case *A*

S	n_s	A_s	$A_s{}^*$
1	174	0·123	0·13
2	44	0·720	0·72
3	10	1·364	1·32
4	8	1·887	1·90

Case *B*

S	n_s	A_s	$A_s{}^*$
1	17	0 112	0·11
2	4	0·740	0·74
3	1	1·342	1·35

Case *C*

S	n_s	A_s	$A_s{}^*$
1	73	0·119	0·14
2	18	0·727	0·75
3	4	1·380	1·38
4	1	1·932	1·99

Table 2. QUADRATIC STRATEGY

Case *D*

S^2	n_s	A_s	$A_s{}^*$
1	65	0·132	0·14
4	20	0·644	0·64
9	3	1·467	1·47

Case *E*

S^2	n_s	A_s	$A_s{}^*$
1	21	0·140	0·14
4	7	0·617	0·63
9	1	1·462	1·46

Case *F*

S^2	n_s	A_s	$A_s{}^*$
1	119	0·109	0·11
4	31	0·693	0 70
9	3	1·708	1·70

Table 3. LOGARITHMIC STRATEGY

Case *G*

log S	n_s	A_s	$A_s{}^*$
0	96	0·093	0·10
0·30	17	0·845	0·84
0·48	6	1·297	1·29

Case *H*

log S	n_s	A_s	$A_s{}^*$
0	32	0·128	0·10
0·30	8	0·730	0·75
0·48	3	1·156	1·13

Case *I*

log S	n_s	A_s	$A_s{}^*$
0	41	0·095	0 07
0·30	7	0·863	0 83
0·48	2	1·407	1 37
0·60	1	1·708	1·70

Table 4. LOG FACTORIAL STRATEGY

Case *J*

log $S!$	n_s	A_s	$A_s{}^*$
0	119	0·194	0·25
0·30	43	0·636	0·65
0·78	20	0·969	1 03
1 38	3	1·792	1·64
2·08	1	2·270	2·35

Case *K*

log $S!$	n_s	A_s	$A_s{}^*$
0	1491	0·177	0 15
0·30	453	0 694	0·48
0·78	232	0 985	1·02
1·38	59	1 530	1·70
2 08	5	2·651	2·50
2·86	1	3·350	3·37

RÉSUMÉ OF SOURCES

Case		
A	Treated chronic schizophrenic	Standard sample.
B	Recovering mild attack of encephalitis	Standard sample.
C	Normal subject.	Spontaneous note
D	Treated chronic schizophrenic, depressed.	Standard sample.
E	Involutional depression.	Standard sample.
F	Depressive psychosis.	Standard sample.
G	Untreated schizophrenia.	Standard sample.
H	Depressive paraphrenia.	Standard samp'e.
I	"The Common Cormorant"	Published nonsense-poem.
J	Epileptic of obsessional personality.	Standard sample.
K	Essay on 'Sense-data' by philosopher.	Whole essay.

FORTHCOMING EVENTS

(*Meetings marked with an asterisk * are open to the public*)

Monday, February 15

Society of Chemical Industry, Pesticides Group (at 14 Belgrave Square, London, S.W.1), at 5.30 p.m.—Dr. T. E. Tomlinson: "Mode of Action of Dipyridyl Quaternary Salts as Herbicides".

Royal Society of Arts (at John Adam Street, Adelphi, London, W.C.1), at 6 p.m.—Dr. Albert Parker: "World and National Energy Resources". (First of three Cantor Lectures on "The Fuel and Power Industries and National Prosperity". Further lectures on February 22 and 29.)

Institute of Metal Finishing (at the Northampton Polytechnic St. John Street, London, E.C.1), at 6.15 p.m.—Mr. T. K. Aiken: "Complexones in Metal Finishing".

Institution of Electrical Engineers, London Graduate and Student Section (at Savoy Place, London, W.C.2), at 6.30 p.m.—Wing Commander E. W. Anderson: "Inertial Navigation Systems".

Tuesday, February 16

University of London (in the Anatomy Theatre, University College, Gower Street, London, W.C.1), at 1.15 p.m.—Dr. K. Ridgway. "Problems of Early Steam Engine Design".*

Institution of Electrical Engineers, Supply Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Discussion on "Soviet Control Engineering Progress" opened by Mr. R. E. Clark.

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. C. H. Gray. "Non-Pancreatic Aspects of Diabetes Mellitus".* (Eleventh of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on February 18, 23, 25 and March 1.)

Society of Visiting Scientists (at 5 Old Burlington Street, London, W.1), at 7.30 p.m.—Discussion Meeting on "Radioactive Fall-out". Chairman: Prof. A. Haddow, F.R.S. Speakers Mr. J. F. Loutit, Dr. W. G. Marley and Prof. K. Mather, F.R.S.

Wednesday, February 17

Physical Society, Colour Group (at the Institute of Ophthalmology, Judd Street, London, W.C.1), at 3.30 p.m.—Dr. J. D. Moreland: "Colour Discrimination, Field Size, Field Position and Acceptor Populations". 4.15 p.m.—Miss J. Cox: "Colour Vision Defects Acquired in Diseases of the Eye".

British Glaciological Society (joint meeting with the Royal Meteorological Society, at 49 Cromwell Road, London, S.W.7), at 5 p.m.—Mr. Herfried Hoinkes (Austria): "Studies in Glacial-Meteorology in the Antarctic, 1957".

Eugenics Society (at Manson House, 26 Portland Place, London, W.1), at 5 p.m.—Mr. Eliot Slater: "Galton's Heritage" (Galton Lecture for 1960).*

Institute of Physics, London and Home Counties Branch (at 47 Belgrave Square, London, S.W.1), at 6 p.m.—Annual General Meeting, followed by Prof. S. Tolansky, F.R.S.: "The Microstructures of Surfaces".

Institution of Electrical Engineers (at Central Hall, Westminster, London, S.W.1), at 6 p.m.—Prof. M. G. Say: "Electrical Machines" (Faraday Lecture).

Society of Chemical Industry, Corrosion Group (at 14 Belgrave Square, London, S.W.1), at 6 p.m.—Dr. Roger Parsons: "Adsorption on Electrodes and Its Relation to the Rate of Electrode Processes".

Royal Institute of Chemistry (at King's College, Strand, London, W.C.2), at 6.30 p.m.—Dr. W. G. S. Parker: "Rocket Propellants".

Thursday, February 18

Royal Society (at Burlington House, Piccadilly, London, W.1), at 4.30 p.m.—Mr. P. Dean: "Vibrational Spectra of Diatomic Chains"; Mr. D. W. Pashley: "A Study of the Deformation and Fracture of Single Crystal Gold Films of High Strength Inside an Electron Microscope".

Institution of Mining and Metallurgy (at the Geological Society, Burlington House, Piccadilly, London, W.1), at 5 p.m.—Dr. A. G. Darnley. "Petrology of Some Rhodesian Copperbelt Orebodies and Associated Rocks".

London Mathematical Society (at the Royal Astronomical Society, Burlington House, Piccadilly, London, W.1), at 5 p.m.—Prof. P. J. Hilton: "Group-like Structures".

Institution of Electrical Engineers, Utilization Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Mr. B. E. A. Vigers and Mr. R. O. Fletcher: "Electricity in the Manufacture of Hydrogen Peroxide".

University College (in the Embryology Theatre, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. E. E. Evans-Pritchard: "Religion and the Anthropologist".* (First of two Special University Lectures in Anthropology. Further lecture on February 25.)

Royal Aeronautical Society (at the Institution of Mechanical Engineers, 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Mr. D. Best: "Missile Control Problems".

British Computer Society, London Branch (at the Northampton College of Advanced Technology, St. John Street, London, E.C.1), at 6.15 p.m.—Mr. R. G. Hitchcock: "The Development of the Use of a Computer for Production Control".

Royal Society of Tropical Medicine and Hygiene (at Manson House, 26 Portland Place, London, W.1), at 7.30 p.m.—Dr. T. F. Davey: "Some Recent Chemotherapeutic Work in Leprosy" with a Discussion on some of the Problems Involved in Clinical Trials.

University of London (at Wye College, Wye, Ashford, Kent), at 8.15 p.m.—Prof. M. M. Swan: "New Light on Growth".*

Friday, February 19

University of London (at the Postgraduate Medical School of London, Ducane Road, London, W.12), at 4 p.m.—Dr. J. A. Fraser Roberts: "Medical Genetics".* (Further lectures on February 26, March 2, 4, 11 and 18.)

Royal Geographical Society (at 1 Kensington Gore, London, S.W.7), at 5.15 p.m.—Symposium on "Space Navigation".

Institute of Physics, Non-Destructive Testing Group (at 47 Belgrave Square, London, S.W.1), at 6 p.m.—Dr. P. Schagen: "Electroluminescent Image Intensifiers".

Society of Chemical Industry, Fine Chemicals Group (at 14 Belgrave Square, London, S.W.1), at 6.30 p.m.—Dr. E. B. Knott: "Organic Chemistry of Colour Photography".

Society for Analytical Chemistry, Microchemistry Group (at the Postgraduate Medical School, Ducane Road, London, W.12, at 6 p.m.—Annual General Meeting. 7 p.m. (joint meeting with the Biological Methods Group)—Scientific Papers.

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Prof. J. Z. Young, F.R.S.: "Eyes, Colours and Shapes".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Lecturer in Health Education, Human Anatomy and Physiology—The Principal, Borough Road College, Isleworth, Middlesex (February 22).

University Demonstrator in the Department of Animal Pathology, School of Veterinary Medicine, to undertake teaching and research in veterinary parasitology—Dr. J. W. Millen, Department of Anatomy, The University, Downing Street, Cambridge (February 26).

Assistant Experimental Officer (with an ordinary degree or diploma in agriculture or botany)—The Secretary, Scottish Plant Breeding Station, Pentlandfield, Roslin, Midlothian (February 27).

Assistant Lecturer or Lecturer in Animal Husbandry in the Faculty of Veterinary Science—The Registrar, The University, Liverpool, quoting Ref. CV/T (February 29).

Lecturer or Assistant Lecturer in the Department of Chemical Engineering, The Houldsworth School of Applied Science, to specialize in separation processes, fluid dynamics or thermodynamics and chemical kinetics—The Registrar, The University, Leeds 2 (February 29).

Physicist or Biophysicist, to assume responsibility for an Electron Microscope—The Secretary, The Institute of Ophthalmology (University of London), Judd Street, London, W.C.1 (February 29).

Research Assistant in the Department of Physics, for work concerned with the fundamental study of electro-luminescence and related processes in single crystal phosphors—The Registrar, The University, Hull (February 29).

Imperial Chemical Industries Research Fellows in Chemistry, Biochemistry, Physics, Engineering, Chemotherapy and related subjects—The Secretary, Academic Council, The Queen's University, Belfast (March 1).

Assistant Lecturer (Terminable) in Botany—The Secretary, Bedford College (University of London), Regent's Park, London, N.W.1 (March 5).

Assistants (2) one in Mathematics and one in Statistics; and a Research Fellow in Mathematics—The Secretary of University Court, The University, Glasgow (March 5).

Lecturer in Radiochemistry—The Registrar, The University, Leeds 2 (March 5).

Lecturer or Assistant Lecturer in Physics—The Secretary and Registrar, The University, Southampton (March 5).

Henry Skynner Senior Student in Astronomy—The Secretary, Balliol College, Oxford (March 12).

Lecturer or Assistant Lecturer (with an interest in applied plant physiology) and a Research Demonstrator in the Department of Agricultural Botany—The Registrar (Room 22, O.R.B.), The University, Reading (March 14).

Lecturer or Assistant Lecturer in Crop Husbandry at Makerere College (University College of East Africa)—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 15).

Milne-Munro Research Fellow (graduate wishing to carry out research in agriculture or cognate subject) at the University College of Rhodesia and Nyasaland—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 15).

Chair of Applied Mathematics in the University of Malaya (Singapore Division)—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 31).

University Demonstrator in Metallurgy—Dr. F. B. Kipping, University Chemical Laboratory, Lensfield Road, Cambridge (March 31).

Senior Lecturer (with appropriate qualifications and experience) in Botany at the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, April 5).

Associated Electrical Industries Fellow for Research in the Department of Physics—The Secretary-General of the Faculties, The Old Schools, The University, Cambridge (April 30).

Chair of Biochemistry in the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, April 30).

Lecturer in Physiology (Histology)—The Secretary of University Court, The University, Glasgow (April 30).

Biochemist (male, with experience of work in the steroid field) at the Sheffield Centre for the Investigation and Treatment of Rheumatic Diseases—The Group Secretary, Sheffield No. 1 Hospital Management Committee, "Lyndhurst", Nether Edge Hospital, Sheffield 11.

Coconut Agronomist (with a university degree in agriculture or botany with experience in the design and analysis of factorial experiments on crops, and preferably experience of experimental work on tropical tree crops) in the British Solomon Islands Protectorate, to supervise existing experiments and to establish a Coconut Research Centre—The Director of Recruitment, Colonial Office, London, S.W.1, quoting Ref. BCD.63/45/04/T.

Lecturer in the Department of Biochemistry—The Secretary, University College, Gower Street, London, W.C.1.

Master to teach Physics and Chemistry in the Sixth Form, with some Scholarship work in one of these subjects—The Headmaster, Blundell's School, Tiverton, Devon.

Senior Technician (male or female)—The Director, Blood Transfusion Service, Roby Street, Manchester 1.

Staines Medical Research Fellow (not exceeding 35 years of age, of English birth, and preferably with a registrable medical qualification), for research in Oxford in one of the medical sciences, such as anatomy, biochemistry, clinical medicine, pathology, pharmacology or physiology—The Rector, Exeter College, Oxford.

Theoretical Physicist, and an Experimental Physicist, for duties which will involve instruction in undergraduate and graduate courses—The Chairman, Department of Physics, Assumption University of Windsor, Windsor, Ontario, Canada.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

Science Books for a School Library. Pp. 54. (London: John Murray, 1959. Published for the Science Masters' Association.) 3*s*. 6*d*. net. [812

Annual Report for 1958 (No. 71) of the Marine Biological Station of the University of Liverpool at Port Erin, Isle of Man. Pp. 58. (Liverpool: Liverpool University Press, 1959.) 5*s*. [812

Scottish Marine Biological Association. Annual Report, 1958–59. Pp. 32. (Millport, Isle of Cumbrae: Scottish Marine Biological Association, 1959.) 4*s*. [812

Annual Report of the Overseas Geological Surveys, 1957–58. By Dr. F. Dixey. Pp. iv+48+7 plates. (London: H.M. Stationery Office, 1959.) 2*s*. 6*d*. net. [812

The Nuffield Foundation. Report for the year ended 31 March, 1959. Pp. xii+201. (London: The Nuffield Foundation, 1959.) [812

Institute of Personnel Management. Occasional Papers No. 14: Operational Research and Personnel Management. Part 1 by Stafford Beer. Part 2 by R. W. Revans. Pp. ii+25. (London: Institute of Personnel Management, 1959.) 3*s*. 6*d*. [812

Henry Wiggin and Co., Ltd. Brightray-Coated Exhaust Valves. Pp. 8. (Birmingham: Henry Wiggin and Co., Ltd., 1959.) [812

An Audit of Academic Performance: a Study of the Prediction of Academic Success in the Queen's University of Belfast. By Michael Forster. Pp. 75. (Belfast: Queen's University, 1959.) 7*s*. 6*d*. [812

British Museum (Natural History). Fossil Mammals of Africa No. 17: Fossil Metacarpals from Swartkrans. By J. R. Napier. Pp. ii+18+2 plates. (London: British Museum (Natural History), 1959). 8*s*. [812

Annual Report of the Advisory Council on Scientific Policy, 1958–59. Pp. iv+14. (Cmnd. 893.) (London: H.M. Stationery Office, 1959.) 1*s*. net. [1112

Ministry of Education. 15 to 18: a Report of the Central Advisory Council for Education (England). Vol 1: (Report). Pp. xxxi+519. (London: H.M. Stationery Office, 1959.) 12*s*. 6*d*. net. [1112

Commonwealth Relations Office. Report of the Commonwealth Education Conference. Pp. 65. (Cmnd. 841.) (London: H.M. Stationery Office, 1959.) 4*s*. net. [1112

Advisory Council on Scientific Policy: Committee on Scientific Manpower. Scientific and Engineering Manpower in Great Britain, 1959. Pp. vi+48. (Cmnd. 902.) (London: H.M. Stationery Office, 1959.) 3*s*. net. [1112

The Nature Conservancy. Report of the Nature Conservancy for the year ended 30th September 1959. Pp. vi+120+14 plates. (London: H.M. Stationery Office, 1959.) 6*s*. 6*d*. net. [1112

European Brewery Convention. Report of the Barley Committee—Trials 1958. Pp. 136. (London: Institute of Brewing, 1959.) [2212

The Institute of Personnel Management. Annual Report 1958–1959. Pp. 27. (London: Institute of Personnel Management, 1959.) [2212

The British Council. Annual Report 1958–1959. Pp. vi+114+9 plates. (London: British Council, 1959.) [2212

Building Research Station Digest, No. 128 (November 1959): Stone Preservatives. Jointing Plasterboard. Pp. 6. (London: H.M. Stationery Office, 1959.) 4*d*. [2212

Forestry Commission Leaflet No. 32: Pine Looper Moth, *Bupalus piniarius*. Pp. 10. (London: H.M. Stationery Office, 1959.) 1*s*. 3*d*. [2212

Hospital Library Services: a Pilot Survey. (Report of an Independent Committee sponsored by King Edward's Hospital Fund for London.) Pp. 56. (London: King Edward's Hospital Fund for London.) 2*s*. 6*d*. [2212

Other Countries

Metropolitan Life Insurance Company. Statistical Bulletin, Vol. 40 (September 1959): Recent Increase in Poliomyelitis. Longevity of Industrial Policyholders in 1958. International Trends in the Birth Rate. Firearm Accidents Costly in Lives. Pp. 12. (New York: Metropolitan Life Insurance Co., 1959.) [812

The Heritage of the Human Brain. By Prof. Charles R. Noback. (James Arthur Lectures on the Evolution of the Human Brain.) Pp. ii+30. (New York: American Museum of Natural History, 1959.) [812

Laboratoire d'Astronomie de Lille. Astronomical News Letter No. 96. Pp. 66. Astronomical News Letter, No. 97. Pp. 64. (Lille: Laboratoire d'Astronomie de Lille, 1959.) [812

East African High Commission. East African Meteorological Department Annual Report for 1957/58. Pp. ix+20+4 plates. (Nairobi: Government Printer, 1959.) Sh. 3/50. [812

Bulletins de la Société Royale de Zoologie d'Anvers. No. 10: Composition du Sang des Singes Anthropoides par Rapport au Sang Humain. Par Dr. Yvan Bereznay. Pp. 57. No 11: Un Cas de Tumeur Melanique chez un Poisson Albinos Protopterus Annectens Owen du Congo. Par Dr. Adolphe Dupont et Dr. Roger Vandaele. Pp. 8. No. 12: Deux Nouveaux Acariens Nasicoles chez un Singe Platyrrhinien *Saimiri sciurea* (L) (Lemurnyssidæ: Sarcoptiformes.) Par A. Fain. Acariens Nasicoles Nouveaux chez un Manchot a' Humboldt et un Perroquet Sud-Americain. Par A. Fain et J. Mortelmans. Sur la Presence d'un Nouvel Halarachnide chez un Manchot Papou (Acarina: Mesostigmata). Par A. Fain et J. Mortelmans. Pp. 27. (Anvers: Société Royale de Zoologie d'Anvers, 1959.) [812

Chemotherapy of Animal Trypanosomiasis: a Résumé of Literature. By Dr. A. K. Sen. Pp. xi+210. (New Delhi: Indian Council of Agricultural Research, 1959.) Rs. 9.50; 15*s*. [812

United States Department of Agriculture. Technical Bulletin No. 1208: Ecological Investigations of the Boll Weevil, Tallulah, Louisiana, 1915–1958. By R. C. Gaines. Pp. ii+20. 15 cents. Leaflet No. 453: How to Control Bed Bugs. Pp. 8. 5 cents. (Washington, D.C.: Government Printing Office, 1959.) [812

United States Department of the Interior: Fish and Wildlife Service. Fishery Bulletin 156: Age and Growth of the Redfish (*Sebastes marinus*) in the Gulf of Maine. By George F. Kelly and Robert S. Wolf. Pp. iv+31. (Washington, D.C.: Government Printing Office, 1959.) 30 cents. [812

Indian Forest Records (New Series). Silviculture. Vol. 10, No. 5: Volume Tables for *Acacia arabica*, Willd. (Mainly applicable to Poona Division, Bombay State.) By S. N. Dabral and M. M. Singh. Pp. 12. (Delhi: Manager of Publications, 1959.) Rs. 1.50; 2*s*. 3*d*. [812

Sveriges Geologiska Undersökning. Oversiktskartor med Beskrivningar. Ser. Ba, No. 15: Beskrivning till Jordartskarta över Uppsalatrakten. Av Björn Järnefors. Pp. 46 med tva planscher och en karta i skalen 1: 20,000. Kr. 5. Ser. Ba, No. 17: Description to Accompany the Map of the Quaternary Deposit of Sweden. By G. Lundqvist. Pp. 116. Kr. 5. Avhandlingar och Uppsatser. Ser. C, No. 560: The Measurement of Rock Pressure in Mines. By Nils Hast. Pp. 183. Ser. C, No. 561: Kvartärgeologisk Forskning i Sverige under ett Sekel. Av G. Lundqvist. Pp. 57. Ser. C, No. 562: Jordskalv i Sverige, 1951–1957. Av K. E. Sahlström och M. Bath. Pp. 19. Ser. C, No. 563: Register över Sveriges Geologiska Undersöknings Publikationer, 1858–1958. Av Ragnar Sandegren. Pp. 330. Kr. 10. Ser. C, No. 564: Rocks and Stratigraphy of the Ledfat Area, Vasterbotten County, Northern Sweden. By Jan Offerberg. Pp. 45+2 plates. Kr. 10. Ser. C, No. 565: C 14-Daterade Tallstubbar fran Fjällen. Av G. Lundqvist. Pp. 21. Kr. 3. Ser. C, No. 566: Fran Nordostlis till Lagbaltisk is en Glacialgeologisk Studie i Sydvästra Skane. Av Hans Möller. Pp. 133. Kr. 9. Ser. C, No. 567: Isstrommar och Isavsmältning i Sydvästra Skanes Backlandskap. Av Kaj Nilsson. Pp. 94+2 plates. Ser. C, No. 569: Förekomsten av Tungmineral i Kaolinen pa Ivö. Av Sven Hjelmqvist. Pp. 13. Kr. 2. Förteckning över Sveriges Geologiska Undersöknings—Publikationer, September 1959. Pp. 37. (Stockholm: Sveriges Geologiska Undersökning, 1958 and 1959.) [812

Arid Zone Hydrology: Recent Developments. By Prof. H. Schoeller. (Arid Zone Research No. 12). Pp. 125. (Paris: United Nations Educational, Scientific and Cultural Organization, 1959.) 12*s*. 6*d*.; 2.50 dollars. [1612

New Zealand: Department of Scientific and Industrial Research. Bulletin 135: Lake Monk Expedition: an Ecological Study in Southern Fiordland. By T. Riney, J. S. Watson, C. Bassett, E. G. Turbott and W. E. Howard. Pp. 75. (Wellington: Government Printer, 1959.) 8*s*. 6*d*. [2212

The West Indies. West Indies Meteorological Service Annual Report, 1958. Pp. 19. (Port-of-Spain, Trinidad: West Indies Meteorological Service, 1959.) [2212

Suomen Geodeettisen Laitoksen Julkaisuja. No. 51: The Results of the Base Extension Nets of the Finnish Primary Triangulation. Pp. 57. (Helsinki: Suomen Geodeettisen Laitoksen Julkaisuja, 1959.) [2212

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COMMONWEALTH EDUCATION AND DEVELOPMENT

IN an adjournment debate on aid to under-developed territories on December 17, Mr. A. E. Cooper said that technical assistance is one of the greatest contributions that Great Britain can make, possibly exceeding in importance what we can do in the way of finance. He referred particularly to the assistance we can give in agriculture, and suggested that we should be more concerned as to the manner in which such technical assistance can be given and the right type of technical colleges provided. Mr. Cooper also believes that there are too many organizations through which assistance can be given, and that the Colombo Plan or similar regional organizations provide the best basis and are more effective than the United Nations. Mr. J. Hall, who stressed also the bearing of a healthy balance of payments on the amount of assistance Britain can give, likewise suggested that our most important contribution is in the field of technical assistance. Here the real difficulty is the supply of technicians and skilled labour available, and he believes that the Government could do more to encourage the training of technicians for service overseas, particularly on Government service. The provision of sufficient qualified teachers, too, is a major difficulty in establishing schools, technical colleges and universities overseas, particularly while service overseas does not rank for pension, and Mr. Hall thinks that the Government could help here by making it possible for such service to count for pension when teachers returned to teach in Britain.

Sir Edward Boyle, Financial Secretary to the Treasury, in welcoming the debate, commented that more interest is being taken in Britain and other Western countries in helping the less developed territories than ever before. Expenditure from United Kingdom public funds on economic and technical assistance to overseas countries and territories has risen from about £75 million in 1957–58 to about £100 million in 1958–59, and is likely to reach £150 million in the current financial year, exclusive of military assistance, emergency expenditure, relief measures and such like, which amount to roughly £30 million a year, while it is estimated that of £300 million a year gross of private investment overseas, about half goes to the less-developed countries. Our bilateral assistance goes mainly and normally to Commonwealth countries: contributions to multilateral organizations go in support of their programmes of assistance to member countries. He believes that there is scope for building up the international institutions in this field, and Sir Edward referred in particular to the Colombo Plan, the International Monetary Fund, the International Finance Corporation and the International Development Association, to the last of which, if acceptable plans come to fruition, the United Kingdom will be subscribing about £50 million. Sir Edward welcomed the reference to agriculture and said that it would be impossible to over-rate the importance to the West of the success of India's second Five Year Plan.

This debate followed a discussion on the same subject initiated by the Marquess of Reading in the House of Lords on December 10 which, however, was concerned chiefly with the Colombo Plan and the countries of South and South-East Asia. Lord Reading himself discussed especially the question of teachers, as well as the importance of greater efforts for more adequate and widespread teaching of English in these countries; he also stressed the importance of the quality of the people who are sent out to render technical and other assistance. Lord Lansdowne, the Joint Parliamentary Under-Secretary of State for Foreign Affairs, just returned from the Colombo Plan Conference in Indonesia, referring particularly to technical assistance, said that up to June 1959 the United Kingdom had provided 2,600 training places in Britain for Colombo Plan students, and 360 United Kingdom experts had taken up assignments in Asia. Moreover, there are now in British universities and training colleges some 40,000 students from overseas, and we have already committed ourselves to £150 million under the Colombo Plan, of which nearly £30 million was spent in the year ended June 1959. Like other speakers in the debate, Lord Lansdowne referred to the need for more generous treatment of those who volunteered for teaching service overseas and for ensuring that their opportunities for promotion are not prejudiced thereby. The Earl of Lucan referred to the bearing on the problem of the new scholarships to be provided under the Commonwealth Scholarships Bill, and Lord Salter stressed the importance of careful choice of appropriate institutions, such as the World Bank, for the provision of capital assistance.

Lord Home, in replying on the debate, suggested that there is no field in which scientific and technical knowledge can pay higher dividends than that of agricultural and farming methods in the Asian countries. Referring to the improvement in recent years in international markets, he agreed on the necessity for stability in the prices of the primary products of Asian countries, and reminded the House that our ability to increase our aid must arise out of the improvement in our balance of payments. It is because of the increase in reserves that Britain has been able to double its subscription to the International Bank, and to increase by 50 per cent its subscription to the International Monetary Fund, while simultaneously its contribution from public funds to economic and technical assistance rose from £75 million in 1957–58 to more than £100 million in 1958–59 and has probably increased by a corresponding amount during the past twelve months. Lord Home agreed as to the importance of technical assistance and of appropriate arrangements being made by local authorities and other employers of teachers to ensure that teachers do not suffer in their careers on return to Britain from service overseas. He believes that, for the time being, regional bodies offer the best means of dealing with the problem, and that the time is ripe for a concerted effort in Western Europe to raise the standard of living in the under-developed countries.

In spite, however, of the reference to the Commonwealth Scholarship Bill, there was no reference in either debate to the Commonwealth Education Conference, although the report of the Conference* dealt specifically with the training of teachers, the supply of teachers for service in countries of the Commonwealth other than their own and to technical education, and specific recommendations are made in the report to deal with these problems. Because the Commonwealth is a new experiment in human relationship, founded on a belief in the worth and dignity of the human individual and a recognition of the value of freedom and co-operative action, education is fundamental to the strength and stability of the Commonwealth, and to social justice and human dignity which must be its inspiration. There is an obligation on those with more highly developed educational facilities to help their fellow members.

The expansion of educational systems in all the countries of the Commonwealth requires new teachers on an unprecedented scale, and all countries, whatever their stage of educational development, need to expand and improve their facilities for teacher training. While some awards made under the Commonwealth Scholarship and Fellowship Plan are likely to be in the field of teacher training, in the long term the training of teachers on the scale required to meet normal needs must be achieved by the efforts of the countries themselves. The Conference Committee which dealt with this subject recommended that the teacher-training assistance provided should be carefully directed to secure the greatest possible benefit in proportion to the resources employed, and that some of the additional places now offered should be reserved for advanced or supplementary courses and only a relatively small proportion used exceptionally for initial training. Some of the funds should be used to enable teacher-training staff in the more advanced countries to assist the development of teacher-training in other countries.

Provision to give experience in training institutions to teacher-training staff from countries with less well-established systems of education is also recommended, and some members of teacher-training staffs in colleges receiving Commonwealth students from other countries should be enabled to visit and gain experience of the countries from which these students are drawn. To overcome the widespread shortage of candidates sufficiently equipped both technically and academically for immediate training as technical teachers, the Committee suggests the provision, where necessary by special courses, of a technical background for young people who have an adequate standard of general education. As a development of major importance, it also recommends that a proportion of the places to be made available should be allocated to training in the teaching of English as a second or later language.

The report stresses the importance of arrangements being made to receive students suitably on arrival and to induct them into their new surroundings and way of life, and of efforts, both before and after their arrival in the host country, to determine whether they need any special instruction in the language of their host country. Countries benefiting from the assistance offered by the United Kingdom should normally bear the cost of travel of their students, but a certain sum should be allocated from United Kingdom funds to assist the travel of students from relatively small and isolated territories. Adequate maintenance allowances are regarded as an integral feature of assistance towards training, and the cost of maintenance should be fully met from any grants made, even if this involves reducing the number of places originally offered by the United Kingdom.

A further Committee of the Commonwealth Education Conference considered the extent to which the countries of the Commonwealth could help each other to meet their needs for training teachers and staff who will train teachers, and the arrangements for providing such assistance. The numbers of primary school teachers required cannot be estimated with accuracy, but clearly many thousands will be required during the next four or five years, mainly from the countries concerned; but to assist these countries to train this large number, there is an immediate need for some 400 or 500 posts to be filled in their teacher-training institutions. Well over 1,000 teachers a year are needed for secondary schools, and about 200 teachers a year for technical schools, while universities throughout the Commonwealth are in need of staff, often in highly specialized subjects. The greatest needs are in Africa.

Already more than 2,500 teachers a year leave the United Kingdom for service in other Commonwealth countries and the Government is to make every effort to increase this number. Canada, Australia and New Zealand made definite offers of assistance at the Conference, and both India and Pakistan hope to encourage their teachers to serve in other Commonwealth countries. In its report the Committee recommended that, to ensure that assistance should be applied at the most fruitful and effective points, governments should concentrate on filling what might be described as key posts in countries where there is a shortage of teachers and that, where appropriate, the filling of such key posts should be facilitated by special allowances to teachers, assessed for the individual appointment. The interests of teachers serving abroad should be safeguarded in such matters as housing, children's education, adequate recognition of teaching service abroad, promotion prospects, re-employment and secondment rights, superannuation and re-settlement on return. The Committee also recommended that the question of establishing a Commonwealth Education Service should be examined, and that a small organization should be established to provide information about vacancies and the availability of staff for service abroad. Steps should also be taken to promote a climate of opinion which will recognize service abroad as a professional asset, and consideration should be given to the secondment of teachers between Commonwealth countries or between regional authorities in one country and another.

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* Commonwealth Relations Office. Report of the Commonwealth Education Conference. Pp. 65. (Omnd. 841.) (London: H.M. Stationery Office, 1959.) 4s. net.

The Committee which considered the extent to which the countries of the Commonwealth could help each other to meet their needs for developing education and training in technical subjects, including training in industry, and the arrangements for providing such assistance, recognized that the training of technologists and technicians in adequate numbers is a serious problem, but pointed to some measures which could be taken immediately. A substantial number of additional places for Commonwealth students will be made available in the universities and technical colleges of the United Kingdom and Canada, and India and Pakistan are willing to offer similar facilities. The Committee strongly recommended that the interchange of scientists and technologists, which is of lasting benefit, should be encouraged and that all countries offering scholarships, other awards or facilities for education, training or research should devote a good proportion of the awards or facilities to technical education. More places should be provided in universities and higher technological institutes in Commonwealth countries for students from small countries which have no such facilities, and the governments of small Commonwealth countries should consider the co-operative establishment of regional technical teacher-training colleges. Postgraduate scholars in science and technology should be encouraged to take part-time teacher-training courses, and the Committee also directs attention to the need for close and continuous attention to the supply of books and periodicals, as well as to the value of consultation with the appropriate professional bodies regarding qualifications awarded by technical institutions so as to ensure a high and uniform standard of attainment.

Normally, arrangements for mutual aid in technical education should be made bilaterally between the countries concerned, and every country should set up a permanent organization to supervise the detailed operation of mutual assistance; but a small information centre established in London might provide information about facilities for technical education and sources of advice available in the Commonwealth. Finally, the Committee recommends that efforts be made to increase mutual assistance by providing opportunities for training and experience in industry from people from Commonwealth countries, and that technical education should receive a full share of all the various kinds of financial and other assistance resulting from the Conference.

The report of the Commonwealth Education Conference estimates that the arrangements agreed at the Conference will involve new expenditure of at least £10 million during the next five years, and it recommends the establishment of a committee in London composed of a representative of each member country of the Commonwealth, with appropriate arrangements to represent the interests of the Colonial territories and provided with a small administrative unit. For the rest, the strength of the new arrangements rests in men and women and the cultivation of personal relationships; and the administrative arrangements proposed are designed to assist the normal channels to flow more freely. Apart indeed from the stimulus given by the Conference to the flow of ideas and exchange of experience, this stimulus to the interchange of scholarship, either in science and technology or in other fields, is of as vital import to the effectiveness of the aid given by the West to the under-developed territories as is the stimulus given to education itself. Both alike should broaden the mutual understanding upon which effective aid must rest and remove suspicions and distrust. Now that the Western governments are seeking to co-ordinate their plans for economic aid to the under-developed countries, education assumes an increasing importance not simply in the matter of technical assistance but also in the establishment of institutions which will enable assistance, whether financial or economic or social, to be given efficiently and swiftly to the greatest effect.

MECHANICS IN THE MIDDLE AGES

The Science of Mechanics in the Middle Ages

By Marshall Clagett. (University of Wisconsin Publications in Medieval Science, Vol. 4.) Pp. xxix + 711. (Madison, Wisconsin: University of Wisconsin Press, 1959.) 8 dollars.

It has long been customary to consider the Middle Ages as a period without any importance for the rise of modern natural science. The Greek contributions were duly studied; then came a blank of several centuries, commonly designated as the Dark Ages, and the story began again with Galileo, whose work seemed to arise out of nothing. The scholastic ideas on natural science were deemed wholly barren and, if ever mentioned, only to be ridiculed.

A marked change in this unfruitful and essentially unhistorical approach was brought about at the beginning of this century by Pierre Duhem, French historian of science, who in a series of brilliant works assumed an entirely new attitude towards the scholastic ideas on natural science in general and on mechanics in particular. He showed that several important notions and results, which always had been considered as typical of the revival of learning in the 16th and 17th centuries, had to be dated back some two or three centuries. His investigations were continued in Europe by C. Michalsky, Anneliese Maier and others. At the same time there arose in the United States a new centre of medieval studies at the University of Wisconsin. There Marshall Clagett notably distinguished himself by penetrating studies in the field of medieval mechanics. The book mentioned above, together with an earlier volume on medieval statics published some years ago by him in collaboration with Ernest Moody ("The Medieval Science of Weights", Madison, Wisconsin, 1952), may be considered (except for new discoveries) the definitive exposition of our present knowledge of the subject. The huge volume is divided into four parts, the first of which contains the essential contents of the former volume with some important supplements. Parts II and III are devoted to kinematics and dynamics respectively and Part IV is an extremely useful summary of the whole subject of medieval mechanics. After an introduction the author generally gives an English translation of the documents at issue, followed by a lucid commentary and by the original text.

The most important subjects treated are: (1) the work of Gerard of Brussels, the importance of which was appreciated some years ago by the author and discussed by him in vol. 12 of *Osiris* (73–175; 1956); (2) the contribution of the scholars of Merton College, Oxford, especially Thomas Bradwardine, William Heytesbury, Richard Swineshead and John of Dumbleton, towards the kinematics of uniformly accelerated motion; (3) the application of plane geometry to kinematics in the graphical method, resulting in a geometrical proof of the so-called Mertonian rule for uniformly accelerated motion, formerly called Oresme's rule by Duhem; (4) Bradwardine's dynamic law of movement, which, though not valid, contributed essentially to the rise of mathematical physics; (5) the theory of impetus and its application in the explanation of the acceleration of free fall and of the movement of celestial bodies; (6) the daily rotation of the Earth, as discussed by Buridan and Oresme, who both advanced strong arguments in favour of it, without, however, bringing themselves to accepting it.

The way in which all these subjects are treated by the author deserves our admiration and assures him a prominent place among the authorities on medieval mechanics. E. J. DIJKSTERHUIS

PHYSIOLOGY OF REPRODUCTION

Reproduction in Domestic Animals

Vol. 2. Edited by H. H. Cole and P. T. Cupps. Pp. xi+451. (New York: Academic Press, Inc.; London: Academic Press, Inc. (London), Ltd., 1959.) 13 dollars.

FORMERLY the physiology of reproduction was studied mainly in small laboratory animals, but, with the increasing economic importance of the larger domestic animals, during the past two decades much work has been done in this field. The second volume of the book on "Reproduction in Domestic Animals" emphasizes the differences which exist between species in the details of their reproductive process.

Six of the eleven chapters, each written by an expert on the subject, deal with the male: spermatogenesis by Ortavent, biochemistry by Mann, libido and storage of semen by Rowson, insemination by Almquist, and the fowl by Lorenz. These present a most comprehensive review of recent literature on the subject. Among many interesting subjects dealt with are calculations of the daily sperm production, species differences in semen characteristics, factors affecting libido and semen quality, the deep-freezing of semen, differences in the technique of insemination in different species, and the effects of œstrogens on the cock. Among the other chapters, one on nutrition by Moustgaard is an important contribution, for among other things it gives a very good account of the nutrient requirements for fœtal development in different species, a subject which has hitherto received but little attention. The effects of under-feeding of the various food constituents, including vitamins and minerals, on the development and viability of the offspring as well as on the functions of the reproductive organs are also described.

In another chapter, by Clegg and Ganong, other environmental factors affecting reproduction in different species are outlined. These include the effects of variation in daylight hours, temperature and humidity together with 'social stimuli', such as that of the sexual display in birds. The physiology of the female fowl is described by Van Tienhoven, and this includes sections dealing with behaviour in mating and broodiness; the effects of different hormones are also reviewed. A chapter on anatomical and physiological factors affecting fertility by Mixner deals with intersexuality and such things as testicular and ovarian hypoplasia, hernias, cystic ovaries, and embryonic mortality. Hart and Osebold describe the infectious diseases of bacterial, virus and protozoan origin.

The book should be most useful to agriculturists and veterinarians, as well as to those engaged in human medicine, for many of the experimental results reviewed are fundamental to the physiology of mammalian reproduction. J. HAMMOND

HISTORY OF BIOLOGY

A History of Biology to about the Year 1900

A General Introduction to the Study of Living Things. By Dr. Charles Singer. Third and revised edition. Pp. xxxvi+580. (London and New York: Abelard-Schuman, Ltd., 1959.) 50*s.* net.

WHEN writing or talking about biology there are two schools of thought, namely the one which maintains that to try to learn or understand the history of the subject is a waste of time, and the other which holds that to understand any subject one must appreciate and know its beginnings and its development. These two differing attitudes can be reconciled by the assumption that biology only found itself as a science about 1850 and what went before is 'irrelevant and immaterial'.

To this reconciliatory suggestion Dr. Singer has produced a stimulating reply in the third and revised edition of "A History of Biology", which is based on the thesis that "the man of science must work backwards for a space . . . for unless we adopt a wide historical outlook we shall find ourselves unable to grasp the nature of the great problems which we are discussing".

After an introduction in which the claims of the historical approach are advanced, the book is divided into three parts. The first part, entitled "The Older Biology", treats aspects of the subject from Hippocrates to Harvey and the circulation of the blood in 1628. In the second section, the "Historical Foundations of Modern Biology" are laid by the development of experimental method in the works of Bacon and Descartes, and followed through the ideas of taxonomy and evolution into the immediate post-Darwin era. There is a number of topics of general interest in the third section such as "Cell and Organism", "Development of the Individual", etc., each of which is examined from a historical point of view.

It is therefore an ambitious book, but as an introduction to the study of living things (which is the sub-title) it seems to me to fall a bit short of the mark. For example, Dr. Singer has chosen his end-point at about 1900. This is sixty years ago, and since that time biology has progressed so quickly and so far that the average reader and television viewer will wonder where he can find the links between this book and the science he sees and hears about him every day. It is realized that to write an up-to-the-minute history of biology is beyond the scope of any man, but when the first edition of this book was published in 1931 it was only thirty years out-of-date, whereas, although additions have been made, it is now sixty years behind the times.

It is proper that a 'history' should contain the names of many people, places and things. But it is bewildering to someone seeking an introduction to find so many things, dates and people on each page and my reaction was to begin to skip certain paragraphs as contributing too little to the main theme.

Nevertheless this book is not simply a compendium and it clearly is a work of scholarship and insight. I found it interesting, stimulating, and in places almost exciting. It is well written and illustrated with much to give to both professional and amateur biologists, and if it has its faults they spring more from the dual task which Dr. Singer has set himself than from any deficiencies in style or material. A. R. GEMMELL

MYCENÆAN AND CLASSICAL GREECE

The Annual of the British School at Athens

No. 52, 1957. Pp. viii+264+50 plates. (London: The British School at Athens, 1959.) 63*s*. net.

THIS is an interesting number. J. Boardman writes on "Early Euboean Pottery and History", and re-opens the question whether the "Chalcidian" ware of the sixth century B.C., assigned by fashionable archæologists to Italiote workshops, was in fact made in Chalcis after all. Euboic letter-forms show, he thinks, close contact between Euboea and Phoenicia in the eighth century B.C. The notorious early war between Chalcis and Eretria for the Lelantine Plain is dated by most scholars around 700 B.C., and supposed to have ended in the triumph of Chalcis. Boardman points out, however, that possession of the plain, necessary to Chalcis, matters little to Eretria, and that Eretria apparently suffered no material setbacks between 800 and 600. I wonder whether one can go farther, and put the end of the war, the Chalcidian victory and the rise of Chalcidian ware all well into the sixth century. This is a very stimulating paper.

A. Andrewes considers early Hellenistic lists of *patrai* in Kamiros. At this date, they were subdivisions of local tribes, and represented a complicated modification of the earlier gentile *patrai*.

V. Karageorghis identifies a Mycenæan vase-painter working in the Levant, who favoured chalices and bowls among shapes, and animal protomae among ornaments.

R. A. Higgins re-examines the Aegina Treasure, now in the British Museum, which recent scholars have dated at about 800 B.C. He demonstrates that, however it reached Aegina, it is mostly, perhaps entirely, Middle Minoan, of about the seventeenth century, and close in style to the treasure of Mallia. His arguments seem conclusive and important.

G. E. Bean and J. M. Cook continue their survey of the Carian Coasts. They retail evidence for their view that classical Cnidus was at Burgaz, near Datça, and that the classical Triopion, at Kumyer near by, had an enormous sea-wall. They also examine the large walled city of Theangela, in their view a creation of Mausolus. But they attribute the large landward wall of Iasus to the late-fifth-century Persian rebel, Amorges, the ally of Athens. Moreover, they believe that at Meropis, on Cos, the Athenians set up their own copy, in Attic lettering, of the famous monetary decree of about 449 B.C., because they had an Athenian station there. The argument is interesting, although we could not have inferred from the decree that coins with the legends "KOION" and "KOS" would soon afterwards be struck at Meropis. Indeed, the wording of the decree seems to give a monopoly of striking to the mint at Athens. This article sturdily and refreshingly rebuts various pedantic dogmas of Louis Robert.

T. E. Jones, L. H. Sackett and C. W. T. Eliot survey the Dĕma, the famous westward-facing wall between the Athenian and Thriasian Plains, and conclude that it dates from the years of panic after Chaeronea (338 B.C.). They note that its plan uses the "indented trace", here closely resembling the landward wall of Iasus, which they date (p. 182) to the fourth century. They were unaware, of course, of Cook and Bean; and it is amusing to see these various dates put forward in one annual.

The number also includes some stray linear *B* tablets from Cnossus, published by John Chadwick, the last harvest of Wace's campaigns at Mycenae, R. Hope Simpson's arguments for a Mycenæan Messene and Sinclair Hood's account of a tower and wall of *c*. 300 B.C. near the Zafer Papoura cemetery at Cnossus. HUGH PLOMMER

BIOCHEMISTRY OF HUMAN GENETICS

Human Biochemical Genetics

By Dr. H. Harris. Pp. viii+310. (Cambridge: At the University Press, 1959.) 37*s*. 6*d*. net.

NO subject in biology is of greater current interest than genetics, and the genetics of human beings is becoming of immense practical importance. It is ironical that most of our knowledge of human genetics comes from a very few fields—in particular blood groups and biochemistry. Several excellent treatises on blood groups are available, but this book by Dr. Harris is the first comprehensive text on the latter field.

In his introduction Prof. L. S. Penrose rightly remarks that the publication of A. E. Garrod's classic "Inborn Errors of Metabolism" marked the beginning of a revolution in genetics. Yet its fruits are slow to ripen. From the last edition of Garrod's book in 1923 until Dr. Harris published his previous work "Introduction to Human Biochemical Genetics", twenty years passed, during which time very little progress was made in human biochemical genetics. Since then a flood of new and fascinating work has appeared—for example, that on the glycogen storage diseases, galactosæmia and the chemical differences between the hæmoglobin variants. The present volume is thus much more than a new edition of Dr. Harris's previous book. It is a good deal longer and well up to date. The chapters deal in succession with a historical sketch, simple genetics, the metabolism of amino-acids and of carbohydrates, the abnormal hæmoglobins, the blood groups, plasma proteins, miscellaneous conditions and the problem of gene action. The last chapter is in many ways the weakest, though it is undoubtedly the most difficult subject to handle. The other chapters are all of an even and high standard. In matters of fact the book is remarkably accurate ; in matters of interpretation it is lucid and well balanced.

The author has somewhat cramped his style by omitting most of the subjects which are speculative, poorly understood, or which cannot be given a firm chemical basis. For example, little mention is made of favism, or of the ability to taste phenylthiocarbamide, or to smell hydrogen cyanide. Population genetics is only briefly treated. This gives the reader the impression of a neatly ordered but restricted field, and the book is therefore less stimulating than it might have been if some of the more provocative themes had been included.

One of the most encouraging aspects of the work described here is the remarkable possibility of cure of the symptoms of some of the inherited diseases. This emphasizes the point that whatever the eugenic problems of our remote descendants, the challenge of our own time is primarily a therapeutic one.

It is a pleasure to handle a book which is so well printed and well bound. The format is the Cambridge University Press's best—well-spaced type on good paper, with contrasting founts for genetic symbols, and clear uncramped figures. The index is reasonably full. It is very well worth the price, and should be in the library of every department of genetics. P. H. A. Sneath

COTTON AND GENETICS

The Application of Genetics to Cotton Improvement

By Sir Joseph Hutchinson. Pp. viii+87. (Cambridge: At the University Press, 1959. Published in association with the Empire Cotton Growing Corporation.) 15*s*. net.

THIS slim volume of eighty-six pages is based on a number of lectures given by Sir Joseph Hutchinson at the North Carolina State College, with some additions from recent work at the Namulonge Station, of which he was director.

The comprehensive title of the book is scarcely applicable to the restricted range of topics discussed. The reader is led to expect a scholarly appraisal of the methods employed and results obtained in other countries and by other workers. He will be disappointed.

The work is divided into six sections, which deal respectively with cotton relatives, the origin and spread of the Old World cottons, the origin and spread of New World cottons, the pattern of evolution in cotton-breeding systems, and the improvement of African cottons.

Discussion of problems implied by the title has to be confined within the limits of forty pages. This must be the author's excuse for a mode of treatment lacking in historical perspective, and which promulgates numerous one-sided opinions on questions still acutely controversial.

Much space is given to a discussion of the theory of the recent origin of the New World tetraploid cottons put forward by the author and his colleagues, R. A. Silow and S. G. Stephens. When first put forward in 1947, this theory was greeted with almost unanimous incredulity. The author now summarizes recent cytological and archæological evidence which at last consigns the theory to respectable burial, although he does not formally reject it himself.

The work of Manning and Sir Joseph Hutchinson on the response of Uganda cotton to selection is described. It is shown that by using a selection index in relatively inbred material, a yield advance of the order of 4 per cent per annum was achieved over eight generations of selection.

This technique is important enough to find a place in cotton breeding, but will probably not come into general use unless it can be combined with the mass pedigree system, and with the recent extensions and modifications of this used by Knight in the Sudan. These comprise selfing alternated with panmixis ; transfer of genes, and transfer of gene complexes by alternate generations of back crossing and selfing.

Nowhere is it stated that the increase in yield obtained in Uganda was in a cotton with the minute yield of 1,000 lb. of seed cotton per acre—approximately one-third of that currently obtained in more favoured countries.

The references to breeding policy in Egypt are unfair and misleading. The brilliant work of Balls and his colleagues in Egypt in almost every branch of science bearing on both yield and quality has been a source of inspiration to cotton workers for nearly a half-century. To omit the name of the inventor of cotton technology is inexcusable ; to misrepresent his policy is both mischievous and irresponsible.

It is clear that Balls was never unmindful of either yield or quality. He has written books on both subjects. He tried to strike a balance between the two ; he "bred to dollars". Although it receives no mention, the work of Hancock, who subjected hundreds of progeny samples to a small-scale spinning test both for the direct evaluation of quality and for an assessment of genetical purity, is of far-reaching importance since it concerns the very future of cotton as an agricultural crop.

Peruvian workers will note with raised eyebrows the statement that Peruvian Tanguis is "fairly primitive by the standard of most cotton growing countries, being a perennial". In point of fact Tanguis has a low node number, and is no more a perennial than Sea Island, Upland or Egyptian cotton. Far from being primitive it is a highly selected and productive cotton much appreciated on the world market.

Apart from these criticisms, and others for which space is lacking, the book contains much valuable information on the development and evolution of cultivated cottons. It is authoritative when the author refers to material well known to him, and it brings up to date the account given by him and his colleagues in their well-known work "The Evolution of *Gossypium*". S. C. Harland

POTASSIUM–ARGON AND RUBIDIUM–STRONTIUM AGES OF SOME GRANITES FROM BRITAIN AND EIRE

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THE development of modern techniques of isotopic age determination makes possible an absolute definition of the geological time-scale. The scale in common use was constructed by Holmes[1,2] and placed the beginning of the Upper Cambrian at 450 million years ago. He was handicapped, however, by uncertainties in the age and stratigraphic assignment of the samples then available ; and subsequent investigators[3–9] and Holmes himself (personal communication) have found it necessary to suggest that the scale should be lengthened.

The most radical revision was that of Mayne *et al.*[8], who suggested that the age of the Upper Cambrian should be nearer 650 million years. This time-scale has already been criticized on general grounds[10,11], but in view of the importance of the revision it was also considered desirable to check some of their results. In an earlier paper[10] it was concluded that the evidence for this extended time-scale rests largely with the measurements made by Mayne *et al.*[8] on British granites. The present article gives some new data obtained for the same rock units by using the isotope dilution technique for argon determinations, and by making independent age determinations by the rubidium–strontium method.

The work at Oxford of Mayne *et al.*[8] began as an investigation, under the direction of Prof. L. R. Wager, into the ages of British granites and related intrusions. In the British Isles, two major sets of granitic intrusions have unusually well-defined stratigraphic relationships connected with the Caledonian and Hercynian earth movements. No subsequent alteration of these intrusions has occurred, with the exception of late-stage magmatic effects[12,13]. The ages obtained by the potassium–argon and rubidium–strontium measurements on the micas of these intrusions should therefore reflect the actual date of intrusion and crystallization of these magmatic bodies.

Among the Caledonian intrusions are the 'granite' masses of Shap, Leinster and Creetown (the latter listed by Mayne *et al.*[8] as "Cairnsmore of Fleet" ; the specimens analysed come from a large granite dyke at Creetown which is probably related to the Cairnsmore of Fleet intrusive) which all cut a folded Ordovician–Silurian series. This series is overlain by Upper Devonian–Lower Carboniferous sediments, which were deposited on the folded and eroded Lower Palæozoic strata.

The adamellite of Shap cuts Ordovician strata ; but the metamorphic aureole surrounding the Shap affects Upper Silurian (Ludlovian) strata, which are the highest beds involved in the pre-intrusion folding[14]. The basal Carboniferous nearby, which is of Lower Dinantian age, contains Shap pebbles[15]. The age of the intrusion is thus post-Ludlovian and pre-Carboniferous. Further, it is probable that it should be assigned to the Downtonian (the transitional Siluro–Devonian stage) or the Lower Devonian on account of general petrological and structural evidence, which, however, is inconclusive.

In Leinster, the upper limit of age is provided by Ordovician deposits and the lower by Upper Devonian. The Creetown granite cuts Upper Silurian strata ; but no lower age limit is definable. In both cases it is generally assumed from petrological similarities that these masses were intruded at the same period of igneous activity as the Shap adamellite.

The Hercynian granites of Britain occur in Devon and Cornwall, and cut a folded Devono–Carboniferous series which is unconformably overlain by continental sediments conventionally referred to the Permian and Trias. The Dartmoor granite cuts a folded series containing Viséan, Namurian and Lower Westphalian fossils[16], though in the immediate neighbourhood of the granite only Viséan fossils are known[17]. Thus, while the Dartmoor granite is almost certainly post-Lower Westphalian, Lower Stephanian strata in Somerset are affected by the same fold system[18] and it is therefore probable that the granites are of latest Stephanian or Permian age. The Land's End granite is correlated with the Dartmoor granite on geochemical and petrological grounds.

The analytical results are given in Table 1 (potassium–argon determinations at Lamont) and Table 2 (rubidium–strontium determinations at Oxford and Harwell). The analyses were all made on samples of biotite separated at Oxford, except for the Leinster granite which was collected by one of us (J. L. K.), with the co-operation of Mr. M. V. O'Brien, director of the Geological Survey of Eire. Samples were further purified by the analysts separately, so that it is not possible to compare analytical details between laboratories. All potassium analyses, however, were made on carefully split aliquots of the final preparations used for the argon measurements.

The isotope dilution method using enriched argon-38 as 'spike' was used for all argon determinations. Over the period occupied by this work. five different spike sets were employed. Analyses of the Massachusetts Institute of Technology standard biotite by four spike sets are given in Table 3. These results agree with measurements reported by four other laboratories on this standard to within 1 per

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Table 1. POTASSIUM–ARGON AGES
$\lambda_\beta = 4{\cdot}72 \times 10^{-10}$ yr.$^{-1}$; $\lambda_e = 0{\cdot}584 \times 10^{-10}$ yr.$^{-1}$

Rock unit and location	Mineral	Specimen number Oxford	Specimen number Lamont	Sample wt. (gm.)	A-38 spike pressure (cm. Hg × 10^4)	A-40* (p.p.m.)	A-40* / Total A-40	Potassium (per cent)	Apparent age (m.y.) This study	Apparent age (m.y.) Mayne *et al.* (ref. 8)
Hercynian Granites										
Dartmoor granite Haytor Rocks, Dartmoor, Devon	Biotite	20,503	1-*B*	2·388	21·0	0·148	0·66	(6·93)	279 ± 8	
			3-*B*	2·088	5·19	0·132	0·94	6·57	263 ± 13	
			4-*B*	3·577	6·09	0·150	0·98	7·22	271 ± 8	
			4-*B*	3·072	6·09	0·148	0·91		269 ± 8	
							Weighted average		271 ± 5	305
Land's End granite Lamorna Cove, Land's End, Cornwall	Biotite	9,437	1-*B*	1·883	5·19	0·134	0·90	7·17	250 ± 15	340
Caledonian Granites										
Shap adamellite Shap, Westmorland	Biotite	7,680	1-*B*	2·568	6·18	0·198	0·91	6·36	393 ± 12	
			1-*B*	2·505	5·19	0·196	0·97		391 ± 20	
		20,405	2-*B*	2·870	6·09	0·185	0·90	5·89	397 ± 12	
			2-*B*	2·429	6·09	0·178	0·85		383 ± 12	
							Weighted average		391 ± 7	475–510
Creetown granite Creetown, Kirkcudbrightshire	Biotite	20,501	2-*B*	1·488	4·91	0·215	0·92	7·18	382 ± 11	470
			2-*B*	1·528	4·91	0·219	0·96		390 ± 12	
								Average	386 ± 8	
Leinster granite Dublin, Eire	Biotite	—	1-*B*	3·622	6·09	0·184	0·86	6·21	377 ± 11	
			1-*B*	3·173	6·09	0·187	0·97		381 ± 11	
	Muscovite	—	1-*M*	2·836	6·09	0·258	0·93	8·32	392 ± 12	
			1-*M*	2·450	6·09	0·258	0·95		392 ± 12	
							Weighted average		386 ± 6	—

* Indicates radiogenic component.
Parentheses indicate potassium analysis on identical material at Oxford, by flame photometer. All others by Ledoux and Co.

cent (P. M. Hurley, personal communication). The coefficient of variation of radiogenic argon measurements at Lamont, based on many duplicate analyses over the past two years, is less than 2 per cent. The absolute uncertainties for samples in this age-range are estimated to be ± 2 per cent, based on the performance of the spike calibration system and the results shown in Table 3. A somewhat higher error is assigned to the samples run with spike ($5{\cdot}19 \times 10^{-4}$ cm. mercury) as this was calibrated indirectly by runs on micas of known radiogenic argon content. The details of these experimental procedures are being given elsewhere[19].

The potassium analyses were performed (with one exception) by Ledoux and Co., Teaneck, New Jersey, using a flame photometric method which has been shown to be reproducible with a coefficient of variation of 1–2 per cent based on blind duplicates. Their results agree with those of the University of Minnesota Rock Analysis Laboratory and isotope dilution analyses at the Lamont Geochemical Laboratory to within 2 per cent. Thus it may be assumed that the individual argon-40/potassium-40 ratios have a coefficient of variation of 3 per cent in this work. Additional inter-laboratory calibrations up to mid-1957 are given by Baadsgaard *et al.*[20] which show a coefficient of variation of only 3–4 per cent on the argon-40/potassium-40 ratios among all contributing laboratories (including Lamont).

The Dartmoor sample 1-*B* was analysed for potassium in the Oxford laboratory by C. McInnes and is believed to be accurate to at least 2 per cent, based on inter-laboratory calibration standards. The argon analysis was done on the identical material. An insufficient quantity was available for analysis by Ledoux and Co.

The potassium–argon isotopic ages were calculated using the decay constants indicated in Table 1. The average age was computed by calculating weights for the individual determinations from their probable errors. The Land's End granite sample was so small that a larger uncertainty is necessarily assigned to the potassium–argon determination of this specimen; but the agreement with the Dartmoor ages is such that the intrusions could have been emplaced at about the same time. The Shap, Creetown and Leinster granites have the same age within the experimental errors. The biotite from the Leinster granite is intimately chloritized and may have suffered slight loss of argon, but the difference between the average muscovite and biotite ages can also be comprehended by experimental error.

Determinations of the age of these micas by the rubidium–strontium method have been made independently at Oxford and at Harwell (Table 2). Both laboratories employed isotope dilution techniques throughout, using enriched rubidium-87 and either strontium-84 or strontium-86 tracers supplied by the Atomic Energy Research Establishment, Harwell. The Harwell techniques have already been described[21]. At Oxford the rubidium and strontium contents, and strontium isotopic composition, of each specimen were determined on a Metropolitan-Vickers *MS 5* solid-source mass spectrometer; analytical details will be discussed in a subsequent paper.

The absolute error in the radiogenic strontium-87/rubidium-87 ratios determined at Oxford is estimated at ± 3 per cent, based on the agreement with the Harwell results and with samples the ages of which were also determined at Lamont (by B. J. G.) and elsewhere. The errors quoted for the Harwell determinations were calculated from the spreads of the various duplicate analyses. A half-life of $(4{\cdot}7 \pm 0{\cdot}1) \times 10^{10}$ years is used for rubidium-87 [22]. The rubidium–strontium ages for the Caledonian (Shap and Creetown) granites agree quite closely with the potassium–argon data reported in Table 1, and again indicate that the two intrusions were contemporaneous. The

Table 2. RUBIDIUM–STRONTIUM AGES OF BIOTITES
$t_{1/2} = 4 \cdot 7 \times 10^{10}$ yr.

Rock unit and location	Specimen number	Rb (p.p.m.)	Normal Sr (p.p.m.)	Sr* (p.p.m.)	Apparent age (m.yr.)	Laboratory
Hercynian Granite Dartmoor granite Haytor Rocks, Dartmoor, Devon	20,503	1,310	10	1·46	267 ± 8	Oxford
		1,298	25	1·54	284 ± 8	Oxford
		1,303	10	1·52	279 ± 8	Oxford
				Average	277 ± 5	
Caledonian granites Creetown granite Creetown, Kirkcudbrightshire	20,501	555	24	0·85	365 ± 11	Oxford
Shap adamellite Shap, Westmorland	7,680	846 848	12	1·27	385 ± 11	Oxford
		766	36	1·19	380 ± 19	Harwell
		801	39	1·30		
		773	34	1·19	364 ± 24	Harwell
		703	34	1·20		
				Weighted average	381 ± 7	

* Indicates radiogenic component.

rubidium–strontium values are much closer to the present potassium–argon ages than to those of Mayne *et al.*[8].

Should the $5 \cdot 0 \times 10^{10}$ year half-life of rubidium-87 (commonly in use at present) be employed, the value found for the average age of these rocks would increase by 6 per cent, and would agree with potassium–argon dates reported in Table 1 to within 2 per cent. The error in the reported rubidium–strontium ages would have to be about 25 per cent for these to agree with those of Mayne *et al.*[8].

The Hercynian data from all sources do not differ as much as the Caledonian dates, but agreement between the potassium–argon and rubidium–strontium measurements reported here is particularly close. Agreement with the Mayne *et al.*[8] dates would necessitate a 10 per cent error in the rubidium–strontium ages given here.

The age data for the Dartmoor granite should be considered in relation to a concordant uraninite age of 290 ± 10 million years for the Cornwall mineralization[23].

On Holmes's Scale *B*[2] the Carboniferous–Permian boundary was 203 million years and the Siluro–Devonian boundary 313 million years. The measurements reported here thus support suggestions for an extension of that scale. A revised time-scale based in part on some of these measurements has been published elsewhere[24]. The discrepancy between the potassium–argon results reported here and those of Mayne *et al.*[8] (which are given in the last column of Table 1) poses an analytical problem which has not yet been solved.

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Table 3. ARGON ANALYSES ON *B*-3208 (MASSACHUSETTS INSTITUTE OF TECHNOLOGY STANDARD BIOTITE)

Sample weight (gm.)	A-40* / Total A-40	A-40* (p.p.m.)	A-38 spike pressure (cm. Hg × 10⁴)
4·166	0·94	0·689	21·0
1·668	0·92	0·687	6·09
1·985	0·97	0·688	6·09
1·759	0·92	0·687	6·01
1·539	0·97	0·692	4·91

* Indicates radiogenic component.

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[1] Holmes, A., "The Age of the Earth" (Benn, London, 1927).
[2] Holmes, A., *Trans. Geol. Soc. Glasgow*, **21**, 117 (1947).
[3] Faul, H., Ann. Rep. Director Geophys. Lab., Carnegie Inst., Wash., 168 (1956).
[4] Hurley, P. M. (Ed.), M.I.T., Fourth Annual Prog. Rep. for 1956–57 (1957).
[5] Folinsbee, R. E., and Baadsgaard, H., Guidebook, Eighth Annual Field Conf. Alberta Soc. Pet. Geol. (August 1958).
[6] Curtis, G. H., Lipson, J., and Evernden, J. F., *Nature*, **178**, 1360 (1956).
[7] Long, L. E., Kulp, J. L., and Eckelmann, F. D., *Amer. J. Sci.*, **257**, 585 (1959).
[8] Mayne, K. I., Lambert, R. St J., and York, D., *Nature*, **183**, 212 (1959).
[9] Davidson, C. F., *Nature*, **184**, 1310 (1959).
[10] Kulp, J. L., Cobb, J. C., Long, L. E., and Miller, D. S., *Nature*, **184**, B.A.62 (1959).
[11] Davidson, C. F., *Nature*, **183**, 768 (1959).
[12] Firman, R. J., *Quart. J. Geol. Soc., London*, **113**, 205 (1957).
[13] Exley, C. S., *Quart. J. Geol. Soc., London*, **114**, 197 (1958).
[14] Harker, A., and Marr, J. E., *Quart. J. Geol. Soc., London*, **47**, 266 (1891).
[15] Capewell, J. G., *Quart. J. Geol. Soc., London*, **111**, 23 (1955).
[16] Dewey, H., "British Regional Geology, South-west England" (H.M.S.O., London, 1948).
[17] Dearman, W. R., and Butcher, N. E., *Proc. Geol. Assoc. England*, **70**, 51 (1959).
[18] Moore, L. R., and Trueman, A. E., *Quart. J. Geol. Soc., London*, **93**, 195 (1937).
[19] Long, L. E., and Kulp, J. L., *Geol. Soc. Amer. Bull.* (in the press).
[20] Baadsgaard, H., Goldich, S. S., Nier, A. O., and Hoffman, J. H., *Trans. Amer. Geophys. Union*, **38**, 539 (1957).
[21] Smales, A. A., and Webster, R. K., *Anal. Chim. Acta*, **18**, 582 (1958). Loveridge, B. A., Webster, R. K., Morgan, J. W., Thomas, A. M., and Smales, A. A., *Anal. Chim. Acta*, **116**, 744 (1959).
[22] Flynn, K. F., and Glendenin, L. E., *Phys. Rev.*, **116**, 744 (1959).
[23] Darnley, A. G., Age Determination Report No. 6, Atomic Energy Division, Geol. Surv. Gt. Brit. (1959).
[24] Kulp, J. L., Proc. Fifth World Petroleum Congress (June 1959).

RELATIONSHIP BETWEEN THE RATE OF FALL-OUT AND THE CONCENTRATION OF STRONTIUM-90 IN HUMAN DIET IN THE UNITED KINGDOM

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IF the long-term consequences of strontium-90 in world-wide fall-out are to be assessed it is essential to determine the degree to which the contamination of human diet depends on the rate of fall-out as opposed to the cumulative total in soil. In an earlier publication it was concluded that the contamination of milk, especially, depended largely on the rate of fall-out[1]. The very tentative nature of the assessment was, however, emphasized. A more precise assessment is now made possible by a more detailed study of the content of strontium-90 in human diet in the United Kingdom which was made in 1958[2] and by additional results of field experiments[3,4].

More than 40 per cent of the milk produced in the United Kingdom is now sampled every two weeks and bulked according to a statistical plan before analysis. During 1958 it was found that the ratio of strontium-89 to strontium-90 in milk during the summer months was more than five times that in the cumulative deposit and at least half of that in the current deposition in rainfall. Because of the shorter half-life of strontium-89 this ratio decreases by a factor of two every seven weeks; thus the effective delay period between the deposition of strontium-90 and its appearance in milk was 1–2 months. Evidence of the importance of recently deposited strontium-90 in determining the contamination of other foods was also obtained.

Although such observations support the conclusion that the contamination of diet is at present largely determined by the quantity of fall-out deposited in the recent past, they do not enable the magnitude of the contribution from the cumulative total in the soil to be assessed. The ratio of strontium-89 to strontium-90 observed in milk is equally compatible with the contrary views that all the strontium-90 in milk was deposited some two months previously, or that it was equally divided between very recent and very old material; other evidence, however, makes the latter conclusion appear unlikely. If the situation is to be adequately interpreted, it is necessary to solve the equation:

$$C = xDp + yDt \qquad (1)$$

where C is the quantity of strontium-90 in the material under investigation, Dp and Dt are respectively the deposition during the life of the plant from which the strontium-90 was derived and the cumulative deposition in the soil, and x and y are proportionality factors.

For reasons previously discussed[1], it has been concluded that the most satisfactory method now available for solving this equation is to compare the observed levels of strontium-90 from world-wide fall-out in food products with experimental measurements of absorption from the soil. Since 65 per cent of the strontium-90 in diet in the United Kingdom comes from milk and milk products[2] the solution of the equation for this material is of primary importance.

Absorption of Strontium-90 from the Soil

A series of field experiments[3,4] has been in progress for three years, to provide information on the extent to which strontium-90 will be absorbed by crops grown under normal agricultural conditions on representative soil types in the southern part of Great Britain; the results are therefore considered to be applicable to England and Wales. Strontium-89 has been applied as a spray to the surface of the soil which is then cultivated to a depth of 4, 7 or 11 in. before crops are sown. Since the application of carrier-free strontium-89 as a fine spray can be regarded as simulating the deposition of radioactive strontium in rain, these experiments should form a reasonably satisfactory basis for assessing the extent to which plants will absorb strontium-90 which is deposited in world-wide fall-out. When ryegrass, or a ryegrass-clover pasture mixture, is grown under these experimental conditions, it is found that absorption is considerably reduced when the soil has been cultivated to the greatest depth. Thus the relationship between the quantity of strontium-90 in soil and the content of plants will change slowly with time as the strontium-90 penetrates more deeply into the soil. Furthermore, it has been observed in some experiments that irrespective of the depth of cultivation the ratio of strontium-90 to calcium in plants may decrease with the passage of time. Two calculations of the relationship between the quantity of strontium-90 in soil and the content of pasture plants have therefore been attempted:

Calculation A: The immediate situation. To estimate the situation likely to occur soon after strontium-90 has been incorporated in the soil, the results have been used for the entire season's crop when land has been cultivated to 4 in. after contamination. The distribution of strontium-90 down the soil profile caused by this method of cultivation is comparable to that now observed from world-wide fall-out in undisturbed soil.

Calculation B: The long-term situation. Deeper penetration of strontium-90 into the soil must be considered. This calculation is therefore based on the average results obtained at the end of the growing season when grass has been grown on land cultivated to 4, 7 and 11 in. after contamination.

Since the extent to which strontium-90 is absorbed from the soil is influenced by the availability of calcium in the soil, the results of the different experiments have been weighted according to the fraction

Table 1. ESTIMATE OF MEAN RATIO OF STRONTIUM-90 TO CALCIUM EXPECTED IN PASTURE GRASSES UNDER FIELD CONDITIONS IN ENGLAND AND WALES FROM SOIL CONTAINING 1 mc./km.^{2}

'Available' soil calcium (m.equiv./100 gm.)		> 15	10–15	5–10	< 5	Country-wide mean
Fraction of total area		0·53	0·19	0·24	0 044	
Experimental results : Mean Sr-90/Ca ratio ($\mu\mu$c./gm.) in ryegrass	Calculation *A*	0·70	1·26	2·72	8·01	
	Calculation *B*	0·49	0·67	1·90	3·75	
Contribution to country-wide mean Sr-90/Ca ratio ($\mu\mu$c./gm.)	Calculation *A*	0·37	0·24	0·65	0·35	1·61
	Calculation *B*	0·26	0·13	0·46	0·17	1 01

Calculation *A*, the situation shortly after contamination ; Calculation *B*, the long-term situation

of the total agricultural land in England and Wales which is of similar calcium status[5]. The country-wide mean values for the ratio of strontium-90 to calcium expected in pasture grass assuming that the soil contains one millicurie of strontium-90 per km.^{2} have been derived (Table 1). The content of plants has been expressed as the ratio of strontium-90 to calcium and not as a concentration or as the percentage absorbed.; as these latter quantities will vary, depending on yield and on the environmental conditions during the growing season, they are not satisfactory bases for the calculation of the resulting levels in milk.

Fraction of the Strontium-90 in Milk which is attributable to Absorption from the Soil

Since the field experiments were carried out in the southern part of Great Britain, attention is here confined to assessing the probable extent to which absorption from soil is responsible for the contamination of milk in England and Wales. It has been shown that the quantity of strontium-90 deposited in different parts of the country varies closely with rainfall and measurements of the content of rainwater have been made[6,7]. The mean cumulative deposition of strontium-90 per km.^{2} of land throughout the country can therefore be derived as 11·5 mc. in the middle of 1958. The analysis of soil samples from representative undisturbed pastures in different parts of the United Kingdom shows that on average approximately 65 per cent of the estimated cumulative deposition remained in the upper 4 in. of soil[2] and it is known that little strontium-90 had penetrated below this level. It therefore appears reasonable to assume that some 65 per cent of the deposition as estimated by measurements of rainfall was accessible to plants in the upper soil layers ; the mean level of strontium-90 per km.^{2} of soil in pastures in England and Wales in July 1958 was therefore 7·5 mc.

It is thus possible from the data in Table 1 to estimate the mean ratio of strontium-90 to calcium expected in pasture plants as a result of absorption from the soil. In calculating the resultant levels in milk, account must be taken of the discrimination factor of 0·10–0·14 against strontium relative to calcium in their passage from the diet of cattle to milk[8], and of the fact that an appreciable fraction of the total diet of cattle is relatively uncontaminated (for example, concentrates) ; this latter fraction has been assumed to be 20 per cent. The ratio of strontium-90 to calcium expected in milk as the result of strontium-90 entering plants from soil has been calculated on this basis and compared with the levels observed in the country-wide survey of milk during 1958 (Table 2). In the calculation it has been assumed that the discrimination factor against strontium relative to calcium is 0·14 ; since the observed range of values is 0·10–0·14 some over-estimate of the contribution of the soil may have been made.

The calculation suggests that at the present time (calculation *A*) about 20 per cent of the strontium-90 in milk is due to absorption from the soil ; furthermore it appears that, if the present cumulative deposit were more thoroughly incorporated in the soil (calculation *B*), it would lead to a level in milk of approximately 12 per cent of that now due to world-wide fall-out. Since the quantity of available calcium in soil varies between different parts of the country, variations in the magnitude of the contribution from soil are to be expected. Separate calculations have therefore been made for the eight zones into which England and Wales have been divided for the study of radioactivity in milk[2]. It appears that the present contribution of absorption from the soil may range from 10 to 33 per cent of the observed levels in milk.

In this assessment no allowance has been made for the effect of future applications of lime to agricultural land. When land is of low calcium status the addition of lime significantly reduces the uptake of strontium-90. Some estimate of this effect can be made from the results of field experiments[3,4], and it has been calculated that future additions of lime at normal agricultural rates to land of low calcium status would reduce the average absorption of strontium-90 from soil throughout the country by approximately 10 per cent.

The proportionality factor x in equation 1 has been estimated (Table 3) by dividing the difference between the observed and calculated values for milk (Table 2) by the rate of fall-out during the year[7] ; y has been estimated by multiplying the country-wide mean levels in herbage (Table 1) by the discriminatory factor in the transfer of strontium and calcium to milk.

Conclusions

The results of the present calculation for the long-term situation may be compared with the earlier assessment based on 1957 data. The two estimates

Table 2. COMPARISON OF THE MEAN LEVEL OF STRONTIUM-90 IN MILK PRODUCED IN ENGLAND AND WALES DURING 1958 WITH THAT EXPECTED IF STRONTIUM-90 WAS ABSORBED ONLY FROM THE CUMULATIVE DEPOSIT IN SOIL

	Ratio Sr-90/Ca $\mu\mu$c./gm	Percentage of observed value
Observed value from world-wide fall-out, 1958	6·84	
Calculated levels for absorption from soil		
Calculation *A*, immediate situation	1·35	20
Calculation *B*, long-term situation	0 84	12

Table 3. Proportionality Factors for the Estimation of Levels of Strontium-90 in Milk by the Equation $C = xDp + yDt$

Proportionality factor	Calculation *A*, immediate situation	Calculation *B*, long-term situation
x, Dependent on rate of deposition ($\mu\mu$c. Sr-90/gm. Ca per mc./km.^{2} deposited per annum)	1 0	1·1
y, Dependent on cumulative deposition ($\mu\mu$c. Sr-90/gm. Ca per mc./km.^{2} cumulative total in the soil)	0 23	0 14

C, Content of milk: $\mu\mu$c. Sr-90/gm. Ca; Dp, rate of deposition: mc./km.^{2}/annum; Dt, cumulative total in the soil: mc./km.^{2}; x and y, proportionality factors.

are not entirely independent since the results for some half the field experiments used in the present calculation provided the basis for assessing absorption from the soil in the earlier study. Confidence in this approach is, however, encouraged by the fact that the values for the proportionality factor y are little different in the two calculations (0·14 and 0·16). The present estimate for proportionality factor x is less than two-thirds of that previously estimated. This is considered to be due partly to removal of strontium-90 from the soil not being taken into account in the earlier calculation and partly to the fact, to which attention has been directed elsewhere[2], that during 1958 the contamination of milk and other agricultural produce did not rise as greatly as the rate of fall-out. It appears probable that this was because the higher rainfall in that year caused a larger fraction of the deposition to be removed from the plants in wash-off.

Although the present calculations are based on the fullest data now available, their imperfections are clearly apparent. The extrapolation to the country-wide situation from a limited number of field experiments is an inescapably inaccurate procedure. Moreover, alternative methods for utilizing the present data are possible though on balance they appear less reasonable. However, two independent lines of evidence, namely the ratios of strontium-89 to strontium-90 in milk and the present calculation, both indicate the importance of recent deposition as opposed to the cumulative total in determining the contamination of milk. While the calculations here presented have been made with respect to the situation in England and Wales only, the general similarity of the ratios of strontium-89 to strontium-90 in milk throughout the United Kingdom[2] suggests that they provide a reasonable guide as to the situation in the whole country.

It has previously been emphasized that local agricultural conditions and climate may cause wide variations in the direct contamination of vegetation[1]; thus the contribution of the cumulative total to the mean level in diet is likely to vary both between different regions and between seasons in any one locality. The present assessment is advanced solely as the best approximation now available for a single country. It is, however, of interest that comparable evidence of the importance of the rate of fall-out in determining the level in diet is available from elsewhere. The ratios of strontium-89 to strontium-90 in milk produced in the United States and in Canada during 1958 have been shown to be similar to those in the United Kingdom[9]. This does not imply that the situations are identical, since the average ratio of strontium-89 to strontium-90 in rain during the same period appears to have been somewhat higher in the United States than in the United Kingdom. However, it is evident that the effective delay period between the deposition of strontium-90 and its appearance in milk was still relatively short; suggestions[10] that the quantity of strontium-90 in milk in North America is determined by the cumulative total in the soil are not compatible with these observations. Finally, it may be noted that recent evidence from a number of sources suggests that the contamination of wheat and rice[2,11,12] and of green vegetables[2] with strontium-90 also depends largely on the extent of the recent deposition.

[1] Russell, R. S., *Nature*, 182, 834 (1958).

[2] Agricultural Research Council Radiobiological Laboratory, Report *ARCRL* 1.

[3] Milbourn, G. M., Ellis, F. B., and Russell, R. S., *J. Nucl. Energy*, A, 10, 116 (1959)

[4] Milbourn, G. M. (in preparation).

[5] Williams, R. (personal communication, 1958).

[6] Stewart, N. G., Osmond, R. G. D., Crooks, R. N., Fisher, E. M. R., and Owers, M. J., U.K. Atomic Energy Authority Report *HP/R* 2790 (1959).

[7] Crooks, R. N., Osmond, R. G. D., Owers, M. J., and Fisher, E. M. R., U.K. Atomic Energy Authority Report *R* 3094 (1959).

[8] Comar, C. L., Russell, R. S., and Wasserman, R. G., *Science*, 126, 485 (1957)

[9] Russell, R. S., "Radioisotopes in the Biosphere". University of Minnesota (in the press).

[10] Kulp, J. L., Schulert, A. R., and Hodges, E. J., *Science*, 129, 1249 (1959).

[11] United States Atomic Energy Commission, Report *H.A.S.L.* 65 (1959)

[12] Tensho, K., Yeh, K., and Mitsui, S., *Soil and Plant Food*, 5, 1 (1959)

PROGRESS OF CANCER RESEARCH

THE thirty-sixth annual report of the British Empire Cancer Campaign* is a massive volume of more than 600 pages dealing with the component reports from the institutes and individual investigators supported by the funds provided by the Campaign—£637,000 was spent on research in 1958. To attempt to summarize the results or even the trends of all the investigations in progress is no easy task; some idea of the current themes can be gained by simply cataloguing the different branches of work which are being pursued at the various institutions (see last year's review in *Nature*, 182, 1547; 1958).

The researches described in the Report can be grouped into three categories: (*a*) clinico-pathology of cancer; (*b*) radiation-therapy and physics and radiobiology; (*c*) carcinogenesis and carcinogens, chemotherapy and the cytology, genetics, biochemistry and demography of cancer.

Only a few of the hundreds of investigations can be referred to in the present review.

The lung cancer–cigarette smoking problem, after the initial impetus given to its solution by the admirable papers of Doll and Hill, has dwindled into attempts to reproduce the postulated carcinogenic effect of cigarette smoking, using mouse and rat tissue, and into the detection or identification of carcinogens in smoke condensates.

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* British Empire Cancer Campaign. Thirty-sixth Annual Report covering the year 1958. Part 1: The Chairman's Statement and the Accounts. Pp. ii+43. Part 2: The Scientific Report of the Researches undertaken by the Central Organization and its Autonomous Councils in the United Kingdom, and by some of its Affiliated Organizations Overseas. Pp. lviii+636. (London: British Empire Cancer Campaign, 1959.)

Blacklock, continuing the studies initiated by the late Sir Ernest Kennaway, has been inoculating fractions of tobacco smoke condensate into the lungs of rats ; these substances were phenols, wax, neutral fractions, carboxylic acids, nornicotyrine, nicotyrine, metanicotine, myosmine, nornicotine, neutral fraction minus hydrocarbons, hydrocarbons and bases minus nicotine. The histological effect induced by tobacco tar in the lungs of rats and guinea pigs has been to induce squamous metaplasia of the bronchial mucosa, sometimes with papillomatous changes.

Peacock, at Glasgow, has studied the incidence of lung adenocarcinoma in series of mice housed in a soot-free room and also in a room exposed to the unfiltered air of central Glasgow ; he concluded that, "there is no evidence to support the idea that atmospheric pollution is a major factor in the ætiology of adenocarcinoma of the lung in C_3Hf mice. Even in animals with no detectable soot in their lungs, tumours occurred at least as frequently as in those whose lungs had obvious deposits of soot". At the same centre polycyclic hydrocarbons present in diesel and petrol exhaust fumes have been identified and assayed: these were, 3 : 4-benzopyrene, 3 : 4-benzofluorene, tetracene (naphthacene), pentaphene, 1 : 2 : 3 : 4-dibenzopyrene, 11 : 12 benzofluoranthene, 1 : 2 : 9 : 10-dibenzotetracene, 1 : 2 : 4 : 5-dibenzopyrene and 1 : 12 : 2 : 3-dibenzoperylene.

Lasnitski, in Cambridge, has been studying the effect of cigarette smoke condensates on organ cultures of (i) human fœtal lung and bronchial tissues and (ii) the bronchi of newly born pigs.

At Leeds, Green and his collaborators are confirming Wynder's results using cigarette smoke condensate applied to mouse skin ; so far as the experiments go, they show that cigarette smoke contains a weak carcinogen.

The effect has been tried of adsorbing a powerful carcinogen (9 : 10-dimethylbenzanthracene) on carbon particles and exposing mice to an atmosphere containing such an artificial soot ; from the development of superficial papillomas it was concluded that the skin apparently elutes the carcinogen from the carbon, but that this elution does not take place in the subcutaneous tissues, nor do the mice breathing air loaded with the carbon-adsorbed carcinogen develop lung cancer. This result could suggest that the carbon, or soot, might conceivably have protective properties by the adsorption of carcinogens.

Among the latest additions to the catalogue of carcinogens is ordinary 'Cellophane', first discovered by Oppenheimer to induce sarcoma in rats after subcutaneous implantation. Horning and Alexander, and also Price, Geffree and Taylor, find that the tumour-producing potency depends very largely on the area of 'Cellophane' inserted under the skin ; pieces of 9 cm.^{2} gave 46 per cent sarcomas, 2·25 cm.^{2} gave 19 per cent sarcomas and 0·56 cm.^{2} gave 0 per cent ; filter paper (2·25 cm.^{2}) or micro cover glasses gave 0 per cent tumours. In Horning and Alexander's experiments with Visking sausage tubing, 2 cm. × 2 cm. gave 60 per cent sarcomas in rats ; 1 cm. × 1 cm., 20 per cent and 0·5 cm. × 0·5 cm., 0 per cent. Oppenheimer, in 1955, had reported that if the 'Cellophane' is first perforated before implantation the incidence of tumours is very considerably reduced. Developments in this field of 'physical carcinogenesis' are awaited with much interest.

The more recent chemotherapeutic agents used for cancer treatment are *nor*-HN2(di-2-chloroethylamine), amino-chlorambucil, melphalan, busulphan (myleran) and glucosaccharo : 1 → 4 : lactone. Unfortunately, these drugs have as yet had only a very limited degree of success, partly perhaps because the therapist usually tries them on advanced cases, and as a result of his caution he is likely to weight the chances against the treatment.

But if the cure of cancer by drugs is still in its infancy, the alternative approach to the disease, namely, prevention, is at least receiving more support. Doll and Hill came to the conclusion that in Britain four-fifths of lung cancers in men are inescapably connected with cigarette smoking : the Report quotes a parallel statement from Oettlé and co-workers who are engaged in demographic studies of cancer in South Africa. These workers write : "The figures provide further evidence of the influence of manner of life on susceptibility to particular cancers and would justify the conclusion that about two-thirds of white (that is, peoples) cancer deaths are inherently preventable by modification of manner of life and environment. The nature of such modification is for the most part not yet defined".

Oettlé *et al.* base their conclusions on the following data : the incidence of cancer in a population of half a million Bantu Africans was studied for a 3-year period and compared with U.S. White, U.S. Negro and Danish populations, age and sex being taken into account. It appears that urbanized Bantus have a total cancer-rate half that of U.S. Whites and Negroes, that the Bantu have a much higher œsophageal and liver cancer-rate (males) and a higher cervix-rate. Rural Bantu have a lower œsophageal and lung cancer-rate, but a higher sinus and foot melanoma-rate compared with urban Bantu.

The Cape Coloured population, which is of mixed White, Hottentot and Malay origin, shows a lower œsophageal and a higher lung and stomach cancer-rate than the Bantu ; the manner of life of the Cape Coloured peoples resembles much more closely "that of Western races and their tumour incidence tends to resemble that of the American negro more than the South African Bantu".

Burrell, at the Cape Bantu Cancer Registry, reports that in the south-eastern regions of the province the natives have a very high and rising incidence of cancer of the œsophagus and that it appears to be more frequent among the well-to-do ; he states, "Thirty-two instances of œsophageal cancer in man and wife have been recorded as well as other combinations of members of the same kraal household. A strong case is developing for environmental carcinogenesis". In the Bantu of the Mozambique region, Prates and colleagues report that liver cancer is exceedingly common whereas carcinoma of the œsophagus is unknown ; skin cancer is very common and the general distribution of sites of cancer is very different from that in the Bantu of Johannesburg and the Transkei.

Another approach to the problem of cancer in Africa is the Kampala Cancer Registry, set up by the British Empire Cancer Campaign at Makerere College to study the incidence of cancer in Uganda. It has now been in operation for several years and is beginning to show practical results outside statistics. A method for the treatment of Kaposi's sarcoma by intra-arterial injection of nitrogen mustard has proved to be a major advance in the treatment of this lesion, which is common in the area. One of the difficulties of the Registry has been the lack of adequate demographic data as a background to its own figures, and its committee appears to have stimulated a pilot survey of three small areas already completed, and of a survey of one whole country proposed as part of the current census of Uganda, which will result in knowledge of the age structure of the population in detail unusual in a tropical country. A similar registry has begun work in Jamaica, but has so far not reached the stage of reporting results. The value of surveys of this type may well be immense. As death-rates from malnutrition and infections decrease, tumours become more important in tropical areas, and it is well to be pre-warned as to what kinds of tumour are likely to become common: they present a very different pattern from those of Great Britain.

The most interesting development in the virus field in cancer in the past few years has been the investigation by Gross, who found that cell-free filtrates of tissues of the strain of mouse (*AKR*) liable to leukæmia, when injected into day-old C_3H mice, induced leukæmia and salivary gland cancer at a later stage. Salaman has now found that cell-free filtrates ('Gradocol') of leukæmic tissue from spontaneous leukæmia in *AKR* mice injected into 17 hr.-old C_3H mice induced 40 tumours in 15 out of 23 surviving mice; 22 were tumours of the salivary gland. The remainder were subcutaneous tumours of the thoracic and abdominal wall and were epithelial in type and closely imitated mammary cancer; a few were sarcomas.

Co-carcinogenesis has been the subject of experiments by Roe; he finds that the bladder as well as the skin of the mouse can be used as tissue for tests. Pellets of paraffin wax, alone or containing croton oil, were implanted in the bladder by the Bonser technique, and urethane was given by stomach tube in some of the experiments. When the bladder wall was examined (*post mortem*) after the tests had been carried on for 40 weeks, carcinomata, some of which were invasive, were found in a proportion of the mice. The yield of tumours, while appreciable, was insufficient for a precise assay; but it was obvious that both urethane and croton oil, alone or in co-carcinogenic combination, can induce papilloma and carcinoma of the mouse bladder epithelium.

As in previous Reports, a great deal of attention is being given at a number of centres to the study of deoxyribonucleic acid in normal and malignant tissue; one of the most interesting advances is described in the experimental work of Harris and his colleagues at Oxford. Along with other workers he has criticized the assumption made in the important researches of Schoenheimer that the components of all the tissues are in dynamic equilibrium; he points out that, "experiments on animals do not distinguish between intracellular turnover and turnover due to simultaneous multiplication and destruction of cells or to secretion of products by cells". Using the macrophage as an example of a non-multiplying cell and (8-^{14}C) adenine as tracer, he has shown that deoxyribonucleic acid is not synthesized in the cell, and no deoxyribonucleic acid adenine or thymidine was involved in turnover; but the ribonucleic acid was found to have a half-life of 2 hr. when it incorporated exogenous labelled adenine. Using tritium-labelled precursors, no transfer of ribonucleic acid from nucleus to cytoplasm could be detected and the half-life of nuclear ribonucleic acid was found to be 2 hr. both in macrophages and in connective tissue cells.

Table 1. PERCENTAGE CONVERSION OF ADDED ^{14}C-LABELLED GLUCOSE AND SUGAR ALCOHOLS TO CARBON-14 DIOXIDE

500 mgm. slices or cells incubated in Krebs–Ringer bicarbonate solution containing 20 mgm. labelled substrate (0·2 μc. ^{14}C) for 3 hr. at 37° C. Results are given ± S.E.M.

Tissue	D-Glucose	D-Sorbitol	D-Arabitol	D-Xylitol
Spleen	5·0±0·6	1·1±0·2	0·17±0·03	1·8
Thymus	3·5	—	0·1	—
Lung	2·6±0·3	0·70±0·03	< 0·05	—
Brain	9·0	0·50	0·11	—
Muscle	4·0	0·30±0·01	< 0·05	—
Lactating mammary gland	20·1	0·65	< 0·05	—
Kidney cortex	12·4±1·9	6·2±1·8	1·3±0·1	16·1±2·4
Liver	4·6±0·4	3·9±0·5	0·87±0·17	4·8±0·4
Fœtal liver	2·6±0·4	2·0±0·1	0·25±0·03	—
DAB hepatoma	4·1±0·7	1·4±0·5	0·15±0·03	2·1±0·1
β-Propiolactone tumour	7·0	0·62	0·12	—
Ascites tumour cells	4·7±0·4	0·87±0·08	0·10	—

It is now more than thirty years since the carbohydrate metabolism of tumours was used by Warburg as a basis for his hypothesis of the characteristics of malignancy; this theme is still a subject for experimentation and controversy. Glock and McLean have adapted the radiocarbon-labelling technique to the study of the glucose metabolism of normal tissues and tumours. Their results are shown in Table 1, and suggest that when rat liver becomes converted to hepatoma by dimethylaminoazobenzene (DAB), some of the characteristics of the carbohydrate metabolism are lost; but in this respect the resulting malignant tissue can scarcely be distinguished from some other normal tissues. Results for the metabolic capacities of the normal tissues (if obtainable), which were the origins of the β-propiolactone-induced tumour and the ascites tumour, would constitute the essential material for adequate comparison.

Physicists become more and more concerned with less in the field of radiation, and a very large number of the reports from physicists working in this field of cancer research are concerned with identifying and measuring amounts of radiation that are considerably less than those met with in the more usual radiological or radiotherapeutic fields. Very keen interest is being taken in environmental radioactivity, and reports of the measurement of the activity from building materials have been supplemented by those dealing with ingested naturally occurring radioactivity. It is reported that Brazil nuts contain more than 20,000 times the activity found in other common foodstuffs. New and more sensitive methods of detection of β- and γ-radiations have been produced, so that it is now possible to detect as little as 20 μμc. cæsium-137 in a single human milk sample. These advances are of great importance in the study of the radiation-levels received by the population in various areas depending on their dietary habits, on the geological nature of the area in which they live and on the type of materials used in the buildings in which they live and work.

The results of experiments carried out to discover the effects of prolonged irradiation at low levels of dosage on mice indicate a premature ageing under such experimental conditions, and this finding confirms the need for such research into the nature of the irreducible natural radioactive background which is present before the addition of increased man-made radioactive contamination into the daily lives of a large proportion of the population.

The radiation dosimetry in bone from bone-seeking radioactive isotopes is being studied in several centres, and practical confirmation is being obtained of the theoretical calculations which have been reported previously. The correlation of the dose received by the sternal marrow during irradiation treatment with the hæmatological findings in such sternal marrow samples is also being studied. Experiments are also reported which are designed to provide further confirmation of Dr. R. H. Mole's observations that the important factor in the production of radiation leukæmia is the dosage-rate rather than the total dose received.

Confirmation of the advantages of radiotherapy with megavoltage radiation is reported from centres all over the world, and the installation and calibration of such equipment necessitate a considerable amount of time being spent on the basic physical measurements which are necessary before the equipment can be put into routine use.

Instruments for the continuous integration and recording of solar erythomal ultra-violet radiation have been designed in Queensland, Australia. This equipment will be used in conjunction with a survey which is now being made of various aspects of the occurrence of skin cancer in an attempt to assess the chief factors causing skin cancer.

Work is also continuing in several centres on the response of tumours to irradiation in atmospheres of various concentrations of oxygen. The radiosensitivity of normal and of malignant cells alike has been found to be under the control of the concentration of dissolved oxygen in the immediate environment of the cell. The administration of oxygen under pressure aims at raising tissue oxygen tension and hence tumour sensitivity by supercharging the plasma with dissolved oxygen.

An interesting application of the Sandberg–Moore technique for detecting tumour cells in venous blood is reported from the Middlesex Hospital, London. In addition to samples of peripheral venous blood and of blood from veins draining the tumour, samples of hepatic vein blood were taken by trans-cardiac catheterization in order to assess the ability of the liver to clear tumour cells from the blood passing through it. The nine cases of gastro-intestinal cancer in which samples were taken from both portal vein and hepatic vein are of particular interest. Tumour cells (certain or suspicious) were seen in eight cases in the portal vein and in only two in the hepatic. Though the numbers are small, they strongly suggest a very substantial filtration effect by the liver. Curiously enough, even in gastro-intestinal tumour, the peripheral blood appears to be more often positive than the hepatic vein, and though the numbers are again too small to be reliable, this would if confirmed suggest in the first place a substantial by-pass of the liver (perhaps by tumour cells passing up the thoracic duct) and in the second place that the lung is a very much poorer filter than the liver.

A group at Mount Vernon Hospital presents interesting findings on the deoxyribonucleic acid content of the nuclei of tumours, using micro-spectrophotometry of Feulgen-stained preparations. Of 100 carcinomata of the cervix uteri, 40 had modal values at or near the normal diploid value, and 32 near the tetraploid value. No clear relation between ploidy and degree of differentiation was noted, though adenocarcinomata had high figures and both of two tumours of mixed histology had very variable nuclear deoxyribonucleic acid with no defined mode. Limited observations on recurrent tumours suggest that high-count tumours are more resistant to radiation and that recurrences have a higher count than the original tumour. In tumours from women, the presence of the sex chromatin (the small body which represents the heterochromatin of the *XX* chromosome pair in man and some other animals) can be roughly correlated with the degree of ploidy, being usually present in diploid and often double in tetraploid tumours.

OBITUARY

Mr. E. W. B. Gill, O.B.E.

Ernest Walter Brudenell Gill died on December 20 at Watlington, Oxon, after a long illness, at the age of seventy-six. Born in London, the son of a canon, educated at Bristol Grammar School and Christ Church, Oxford, where he was an Open College Exhibitioner in mathematics and later a Scholar, he took a first-class in physics in 1905, a B.Sc. degree in 1909, became University demonstrator in the Electrical Laboratory under Townsend, and a Fellow (1910) and later the domestic bursar of Merton College, Oxford.

During the First World War he served in the Royal Garrison Artillery and the Royal Engineers Wireless Intelligence, which took him to the Middle East. Afterwards he wrote a witty little book, "War, Wireless and Wangles". During the Second World War he again was in Intelligence and operational research. He was a co-inventor of 'window', metal foils dropped from planes so that our aircraft could not be located by enemy radar; he also worked on devices for guiding tanks. After the War he and an American representative interviewed the German scientists. He was surprised to find how little they had contributed to the war effort, some pretending that this was to save the world from destruction. A humorous account of his experiences, intended as a book, remains in the manuscript stage.

Gill's numerous scientific contributions cover a wide range of physical problems. With Pidduck he measured the ionization coefficient in helium. He discovered the effect of the electric field on the saturation current of tungsten emitters in valves and developed an ingenious method of determining the high-frequency conductivity and dielectric constant of the soil by measuring the ratio of the vertical and horizontal components of electric field and the phase angle between them. He investigated extensively electrodeless discharges in high-frequency electric fields, and observed that at very low gas pressure the starting potential becomes very small when a magnetic field applied perpendicular to it is of such value that electrons move through their orbit in a time equal to their periodic time (1937), an effect recently spectacularly demonstrated by Cutolo in Naples, who started a discharge a long distance from the transmitter of an electromagnetic beam. With me he found that in zero magnetic field the initial multiplication of charges at low pressure is due to secondary electrons released from the walls of the glass-vessel. He contributed greatly to our knowledge of static electrification, showed that frictional electricity is caused by contact potentials and found that by freezing-in charges in ice surrounded by water, potential differences of several 100 volts appear across the ice. Later he turned his fertile mind to the elucidation of the mechanism of the rise of sap in trees.

Gill was an outstanding Englishman, a gay character, modest, with an inborn sense of humour and a fund of excellent stories. He had the original mind of a young man coupled with the neat approach of a professional soldier. Debunking ("rebuking sin") was his hobby. Because of his independent judgment, his unfailing logic and his devastating remarks (often noted by the Press), he stepped quite often on people's toes. He never received the recognition he deserved either as a scientist or for his work in the administration of the University or as an Alderman of the City. He was a trusted friend and a devoted husband, who is survived by his wife, three sons and one daughter.

A. von Engel

NEWS and VIEWS

National Agricultural Advisory Service: W. Morley Davies

Mr. W. Morley Davies has retired from his post of senior advisory officer in the National Agricultural Advisory Service. He attended the University of Birmingham, taking a degree in chemistry and botany. During the First World War he was engaged on the manufacture of explosives in British Dyestuffs, Ltd., Huddersfield. Afterwards, he went to Gonville and Caius College, Cambridge, as a research student and worked in the School of Agriculture under Prof. T. B. Wood and Arthur Amos, taking a research B.A. in 1922. At the same time, he also obtained the agricultural diploma. In September 1922 he was appointed head of the Department of Chemistry at the Harper Adams Agricultural College, and later, in 1924, was also appointed as advisory agricultural chemist to the West Midland Province. During that period, mainly in collaboration with the late Prof. G. W. Robinson, Prof. T. Wallace, Sir William Ogg and others, many of the methods now in use in the Soil Survey were developed. In 1946 he was appointed the first director of the East Midland Province in the newly constituted National Agricultural Advisory Service. He was appointed senior education and advisory officer in charge of all the science sections of the Service and stationed at headquarters in London in 1949. As chairman of the Land Restoration Committee and also of the Technical Advisory Conference on the Restoration of Opencast Coal Land, he piloted much work on the technical problems of restoring land to agriculture after it had been subjected to mineral working. He was also a member of the Ironstone Advisory Committee appointed by the Ministry of Housing and Local Government. During the period at headquarters he was secretary of the Agricultural Improvement Council (England and Wales) and assessor for the Ministry of Agriculture on several of the committees of the Agricultural Research Council as well as on the Geological Survey Board.

Dr. N. H. Pizer

Dr. N. H. Pizer, who will succeed Mr. Morley Davies, has been regional soil chemist in the Eastern Region of Great Britain since 1946 and has been interested in soil conditions affecting root development and nutrition of crops, particularly those related to soil conformation and structure. Physical defects such as compacted or structureless layers have been found to occur in many soils and to impede drainage, root development and root activity. Some are due to treatment of the soil and others are natural, and their effects on crops vary with season. Methods of dealing with these defects have been worked out with the farmers concerned. Some defects have required drainage operations or cultivations to a suitable depth when the soil was dry, and others a combination of the two. Dr. Pizer has also worked on the amount and distribution of organic matter in soils under different systems of husbandry and the effects which very low levels of organic matter may have on the physical condition of soils and their ease of working. Another of his interests has been the occurrence of trace-element deficiencies in crops and the development of methods of recognition and treatment. As a result of this work, copper deficiency was found to occur in cereals growing on many fen peat soils and on very light sandy soils previously reclaimed from heath. Yields of cereals were increased from a few cwt. an acre to more than 30 cwt. by the application of ¼–1 lb. of copper per acre as a foliar spray.

After the sea floods of 1953 which affected most of the coast-line of eastern England and extended inland up the river valleys, considerable work was done in the field and laboratory on methods of restoration of soils. Much variation was found between soils in the effects of salt on their physical condition, some being extensively damaged and others not at all, and these variations had an important influence on the treatment that was recommended to restore the land to its former condition. More recently, the problem of magnesium deficiency in fruit crops, glasshouse crops and arable crops on light land has been studied and practical methods of treatment developed.

Paul Instrument Fund: Awards

Awards of grants by the Paul Instrument Fund Committee have been made as follows: £500, in supplement of a previous grant, to Dr. B. V. Bowden, principal of the Manchester College of Science and Technology, for the development of narrow pass-band dye filters; £545, in supplement of a previous grant, to Dr. R. Feinberg, of the Department of Electrical Engineering, Manchester College of Science and Technology, for the construction of an instrument for measuring the intensity of soft X-radiation generated in experimental high-voltage valves; £3,000 to Prof.

D. Gabor, professor of applied electron physics in the Imperial College of Science and Technology, London, and Dr. D. Jones, of the Department of Electrical Engineering in the Imperial College, for the development of an electron interference microscope; £8,100 to Dr. H. H. Hopkins, reader in optics, Department of Physics, Imperial College of Science and Technology, London, for the development of methods of drawing and coating fine glass fibres, and their alignment in ordered bundles for the transmission of optical images; £3,000, in supplement of a previous grant, to Prof. R. O. Redman, professor of astrophysics in the University of Cambridge, for the construction and testing of a thin astronomical mirror of plate glass and of a new type of support system; £6,000, in supplement of a previous grant, to Dr. C. N. Smyth, of University College Hospital Medical School, London, and Mr. F. Y. Poynton, of Northampton College of Advanced Technology, London, for the construction of an ultrasonic microscope.

The Paul Instrument Fund Committee, composed of representatives of the Royal Society, the Physical Society, the Institute of Physics and the Institution of Electrical Engineers, was set up in 1945 "to receive applications from British subjects who are research workers in Great Britain for grants for the design, construction and maintenance of novel, unusual or much improved types of physical instruments and apparatus for investigations in pure or applied physical science".

Commonwealth Scholarship Commission in the United Kingdom

The following have been appointed members of the Commonwealth Scholarship Commission in the United Kingdom by the Secretary of State for Commonwealth Relations, Lord Home, and the Secretary of State for the Colonies, Mr. Ian Macleod: The Earl of Scarbrough, the Lord Chamberlain (*chairman*); Dr. R. S. Aitken, vice-chancellor of the University of Birmingham; Sir David Anderson, recently director of the Royal Technical College, Glasgow; Sir Eric Ashby, master of Clare College, Cambridge; Sir Eric Coates, Finance Minister of the Governor General of India's Executive Council, 1946; Lord Geddes of Epsom, member of the London Transport Executive; Sir Hector Hetherington, principal and vice-chancellor of the University of Glasgow; Sir Francis Hill, chairman, General Purposes Committee of the Association of Municipal Corporations; Sir Cyril Hinshelwood, Dr. Lee's professor of chemistry in the University of Oxford, and president of the Royal Society; Sir Douglas Logan, principal of the University of London; Lady Ogilvie, principal of St. Anne's College, Oxford; Sir Julian Pode, managing director of the Steel Company of Wales, and chairman of the Managing Committee of the United Kingdom for the Athlone Fellowships scheme; Sir Paul Sinker, director general of the British Council, and First Civil Service Commissioner, 1951–54; Dr. R. W. West, principal of Battersea College of Technology. The Commission has been set up under the Commonwealth Scholarships Act recently passed by Parliament to administer the Commonwealth Scholarship and Fellowship Plan in Great Britain.

A Hundred Years of Optics

In 1860 John Henry Dallmeyer took out a patent and issued his first Triple Achromatic lens, marking the foundation to the House of Dallmeyer. J. H. Dallmeyer was born in Westphalia in 1830 and went to Great Britain in 1851. He first set up a business designing and manufacturing lenses and optical instruments at 19 Bloomsbury Street, London. In 1861 he was elected a Fellow of the Royal Astronomical Society, and four years later invented the Dallmeyer Rectilinear lens. Improvements in optical technology brought about the series of *f*/3, *f*/4 and *f*/6 Patent Portrait lenses. The management of the business gradually devolved upon Thomas Rudolph Dallmeyer, who, like his father, possessed a wealth of knowledge in optics. In 1891 the first practically useful telephoto lens was designed and issued by him, only to be followed by the Adon telephoto lens in 1892. One of the earliest treatises on the use of telephoto lenses was to his credit, and "Telephotography" was published in 1899. T. R. Dallmeyer died in 1906, and it was only due to the fact that a private limited company had been formed in 1892 that the business remained established. Later developments included the pioneering of the first fixed-separation telephoto lens, the inventions of the Dallon Fixed-separation Telephoto lens and the Pentac anastigmat lens. It was the Pentac lens that created new standards in large-aperture anastigmal lenses, and which was used during the Second World War for aerial reconnaissance both by day and night. It is with much credit to its pioneers that the House of Dallmeyer celebrates its centenary.

Public Health Laboratory Service in Britain

The Public Health Laboratory Service Bill, which received its second reading in the House of Lords on February 4, transfers the administration of the Public Health Laboratory Service in Britain from the Medical Research Council to a new Public Health Laboratory Service Board appointed by the Minister of Health and subject to his directions. The Board will consist of a chairman and nine other members, one of whom must be medically qualified; two members will be appointed after consultation with the Medical Research Council, two must have had experience as bacteriologists, two will be medical officers of health of local authorities, and two with administrative experience. Clauses in the Bill transfer the staff of the Public Health Laboratory Service from the Medical Research Council to the new Board, and property held for the Public Health Laboratory Service is transferred to the Minister of Health. In introducing the Bill, the Minister for Science, Lord Hailsham, explained that the Service had its origin in an emergency service created shortly before the Second World War at the request of the Committee of Imperial Defence, in order to provide a defence against possible enemy use of bacteriological warfare and against any major outbreaks of epidemic diseases arising from war-time conditions. In the event it was never used to deal with any such major emergency, but it proved a very valuable aid to the medical officers of health in their ordinary work, in view of the inadequacy of the arrangements for bacteriological work for public health purposes. Unlike the Hospital Laboratory Services, it was organized on a national and not on a regional or hospital basis, and consists at present of the Central Public Health Laboratory, eight regional and 51 area laboratories, with several special reference libraries; of the staff of about 1,068, 126 are medically qualified. £1,212,000 was provided for the cost of the Service in the Civil Estimates for 1959–60 and another £50,000 for capital expenditure on buildings. Replying to Lord Taylor, who paid tribute to the immense amount of research carried out by the Service (estimated at about one-quarter of its work), Lord Hailsham readily gave the assurance requested that its research activities would not be curtailed.

British Overseas Information Services

In the House of Commons on February 1, the Chancellor of the Duchy of Lancaster, Dr. C. Hill, gave the estimated cost of the overseas information services during 1959–60 as £16,966,140, of which 30·3 per cent was attributable to the British Council, 39 per cent to the British Broadcasting Corporation's External Services, 18·3 per cent to the Foreign Office, 8·4 per cent to the Commonwealth Relations Office, and 3·7 per cent to the Colonial Office. There were 117 United Kingdom information posts overseas, of which nine were staffed by one United Kingdom-based officer only, and three by one locally-recruited officer only.

Research and Industry in Great Britain

In reply to a question in the House of Commons on February 4, the Minister of Education, Sir David Eccles, as representing the Minister for Science, said that the research needs of the printing machinery, textile machinery and shipbuilding industries were among those being considered for special study by the Department of Scientific and Industrial Research. Since the full co-operation of the firms concerned was essential, an inquiry on a confidential basis was proposed. The report would afterwards be discussed by the industry and steps taken to remedy any defects.

Courses on Nuclear Energy for Engineers and Technical Industrialists

Two international courses on nuclear energy, designed for senior engineers and technical industrialists, are being arranged this summer by the European Nuclear Energy Agency of the Organization for European Economic Co-operation. One course will be at the Centre d'Etudes Nucléaires at Saclay (France) and the other at the Atomic Energy Research Establishment, Harwell. Each will be open to some fifty participants from countries belonging to the Organization, and will cover the technical, economic and industrial aspects of nuclear energy. The course at Saclay (in French) will be held during June 27–July 8; the Harwell course (in English) during July 4–15. The programme in each case will comprise lectures, practical work and visits to installations, and will be presented throughout from the point of view of the engineer and the industrialist. Further information can be obtained from O.E.E.C. European Nuclear Energy Agency, 38 Boulevard Suchet, Paris 16.

Courses in Advanced Technology

The wide range of part-time courses dealing with advanced aspects of technology provided by technical institutions in the London area is described in a bulletin prepared by the London and Home Counties Regional Advisory Council for Technological Education. At Balham and Tooting College of Commerce experts from foreign embassies were invited to discuss the problems of British export policy. A course on energy approximations in structural design was arranged at Battersea College of Technology, while at the Borough Polytechnic bakery sales manageresses were invited to learn how better to sell bakery products. Joinery estimating was available for joinery managers and shop foremen at the Brixton School of Building, and new developments in gas chromatography were available at the Brunel College of Technology. At the City of London College senior executives of tanker operating firms were invited to a series of lectures on the economics of tanker shipping operations. Further details of the bulletin can be obtained from the Secretary, Regional Advisory Council, Tavistock House South, Tavistock Square, London, W.C.1.

The Leopoldina German Academy of Sciences: Elections

The Deutsche Akademie der Naturforscher Leopoldina, Halle, recently elected the following new members, in the sections indicated: *Zoology*, Prof G. P. Baerends (Haren (Gron.) Holland), Prof. Willi Hennig (Berlin), Prof. Werner Jacobs (Munich), Dr. Martin Lindauer (Munich), Prof. Hans J. Müller (Quedlinburg), Dr. Johann Schwartzkopff (Munich), Prof. Niko Tinbergen (Oxford); *Physical Chemistry*, Prof. Gustav Kortüm (Tubingen), Prof. Werner O. Köster (Stuttgart); *Internal Medicine*, Prof. Ferdinand Hoff (Frankfurt/M.), Prof. Hugo Wilhelm Knipping (Cologne), Prof. Ernst von Lauda (Vienna), Prof. Paul Martini (Bonn), Prof. Helmuth Reinwein (Kiel), Prof. Rudolf Schoen (Göttingen).

Clough Memorial (Geological) Research Fund

The Clough Memorial Research Fund of the Edinburgh Geological Society is available for geological research in Scotland and the North of England (Northumberland, Durham, Cumberland, Westmorland and Yorkshire). Grants for the period April 1, 1960–March 31, 1961, will be made shortly, and applications, stating the nature of the proposed research, should be sent before March 31 to the Secretary, Clough Memorial Research Fund Committee, Edinburgh Geological Society, Synod Hall, Castle Terrace, Edinburgh 1.

University News:

London

Dr. H. O. Schild, reader in pharmacology at University College, has been appointed to the University chair of pharmacology tenable at that College, as from October 1, 1961; and Dr. W. F. Widdas, reader in physiology at King's College, has been appointed to the University chair of physiology tenable at Bedford College, as from October 1, 1960. The following appointments are also announced: Dr. H. Lehmann, senior lecturer in chemical pathology at St. Bartholomew's Hospital Medical College, to the University readership in chemical pathology tenable at that College; and the title of reader in chemical pathology in the University of London on Dr. G. R. Webster in respect of his post at Guy's Hospital Medical School.

Oxford

It is announced that the Forestry Commission is to make a grant not exceeding £4,000 for two years beginning April 1, 1960, for the continuation of an investigation into the costs of private forestry and economic information concerning private forestry estates in England and Wales, being carried out in the Department of Forestry under the direction of J. J. MacGregor.

The Night Sky in March

Full moon occurs on Mar. 13d. 08h. 26m. U.T., and new moon on Mar. 27d. 07h. 38m. The following conjunctions with the Moon take place: Mar. 5d. 00h., Aldebaran 0·4° S.; Mar. 20d. 11h., Jupiter 5° S.; Mar. 21d. 14h., Saturn 4° S.; Mar. 24d. 04h., Mars 4° S.; Mar. 25d. 12h., Mercury 0·7° S.; Mar. 25d 12h., Venus 2° S. In addition to these conjunctions with the Moon, Mercury is in conjunction with Venus on Mar. 25d. 09h., Mercury being 1·9° N. There will be a total eclipse of the Moon on March 13 and a partial eclipse of the Sun on March 27, both eclipses being invisible at Greenwich. Mercury can be seen during the first few days of the month as an evening star, but conditions rapidly become unfavourable, and after March 5 Mercury will be too close to the Sun for easy observation. There is an occultation of Mercury by the Moon on March 25, visible from Greenwich and lasting from 11h. 51·1m. to 13h. 03·6m. Venus and Mars are too close to the Sun for easy observation. Jupiter is a morning star, rising at 3h. 30m, 2h. 40m. and 1h. 45m. on March 1, 15 and 31, respectively. It may be seen in the south-east before sunrise; it is in Sagittarius. Its stellar magnitude is $-1 \cdot 7$ and its distance from the Earth in the middle of the month is 490 million miles. Saturn is a morning star, rising at 4h. 30m., 3h. 40m. and 2h. 40m. on March 1, 15 and 31, respectively. It is visible in the south-east before sunrise, and, like Jupiter, is in Sagittarius. Its stellar magnitude is $+0 \cdot 8$ and its distance from the Earth on March 15 is 960 million miles. Occultations of stars brighter than magnitude 6 are as follows, observations being made at Greenwich: Mar. 4d. 20h. 40·6m., θ^1 Tau. (*D*); Mar. 4d. 20h. 44·2m., 75 Tau. (*D*); Mar. 4d. 21h. 40·3m., 264 B. Tau. (*D*); Mar. 5d. 00h. 00·2m., 111 Tau. (*D*); Mar. 11d. 3h. 18·0m., o Leo. (*D*); *D* refers to disappearance. The vernal equinox occurs on March 20d. 15h.

Announcements

A decision by the U.S.S.R. to join the International Federation of Computer Societies brings the number of nations which have ratified the statutes of this new international organization up to twelve. The Academy of Sciences of the U.S.S.R. will be represented in the Federation by its Computing Centre.

An international symposium on Growth: Molecule, Cell and Organism will be held at Purdue University, Lafayette, Indiana, during June 16–18. Further information can be obtained from Prof. M. X. Zarrow, Department of Biological Sciences, Purdue University.

The annual International Starch Conference is to be held at the West German Starch Research Station, Detmold, during April 20–22. Further information can be obtained from W. Kempf, Cereal Research Station, P.O. Box 23, Detmold, West Germany.

A conference on the Computing Laboratory in the Technical College is to be held during May 27–28 at the Hatfield Technical College, which is sponsoring it. Further details can be obtained from W. A. J. Chapman, Department of Mathematics, Hatfield Technical College, Hertfordshire.

The Chemical Society is organizing a symposium on Physical, Chemical and Biological Methods of Study in High Molecular Weight Carbohydrates, to be held in Edinburgh during July 12–14, 1960. Further information can be obtained from the General Secretary, Chemical Society, Burlington House, London, W.1.

The fifth International Congress on Nutrition will be held in Washington, D.C., during September 1–7, under the auspices of the International Union of Nutritional Sciences. Dr. C. Glen King (The Nutrition Foundation, New York City) will preside over the Congress. Further information can be obtained from Dr. Milton O. Lee, 9650 Wisconsin Avenue, Washington 14, D.C.

The general programme of the Madrid Sectional Meeting of the World Power Conference during June 5–9 has now been published. British subjects wishing to obtain further information should write to the Secretary, British National Committee, 201–2 Grand Buildings, Trafalgar Square, London, W.C.2, from whom copies of the programme can also be obtained.

A conference on Solid State Microwave Amplifiers is being organized by the Electronics Group of the Institute of Physics, in conjunction with the Radio Spectroscopy Group, at the University of Nottingham during April 6–8. Further details can be obtained from the Secretary, Institute of Physics, 47 Belgrave Square, London, S.W.1.

The International Medical Congress, Ltd., which has a strong affiliation with the National Foundation of New York, is holding its first International Conference on Congenital Malformations in London during July 18–22. Further information can be obtained from the Executive Secretary-Treasurer, First International Medical Conference on Congenital Malformation, 67 New Bond Street, London, W.1.

The Pesticides Group of the Society of Chemical Industry is organizing a symposium on Fungicides in Agriculture and Horticulture in the spring of 1961. It would be appreciated if workers on these subjects who may wish to submit a contribution to this symposium would get in touch with the hon. secretary of the Pesticides Group, Dr. B. J. Heywood, c/o May and Baker, Ltd., Dagenham, Essex, giving a very brief indication of the scope they would wish to cover.

Prof. R. A. McCance and the Department of Experimental Medicine, Cambridge, are arranging a course on the Clinical Aspects of Nutrition, to be given in the Department of Geology, Cambridge, during April 6–9. The course is open to university and hospital medical and scientific staffs and registered medical practitioners. Further information can be obtained from the Secretary, The Medical School, Tennis Court Road, Cambridge.

The second European symposium on Chemical Reaction Engineering is to be held at the Koninklijk Instituut voor de Tropen, Amsterdam, during April 28–30. The symposium is being organized under the auspices of the European Federation for Chemical Engineering by the two Dutch member societies. Further information can be obtained from P. J. Hoftyzer, Central Laboratory, Staatsmijnen in Limburg, Geleen (L), Netherlands.

Erratum. We regret that in the communication entitled "Shape of the Precipitate in Immunoelectrophoresis", by Dr. Jan Hirschfeld, in *Nature* of January 16, p. 164, Fig. 4 has been printed upside down.

THE INTERNATIONAL COMMITTEE ON LABORATORY ANIMALS

By Dr. W. LANE-PETTER

Laboratory Animals Centre, Carshalton, Surrey

IN December 1956, the International Committee on Laboratory Animals was formed, and a report of this event appeared two months later in *Nature* (179, 240; 1957), describing its background, composition and purpose. After three years, it is timely to review its work and achievements.

The Committee is to-day supported, by direct grant and by representation, by the Council for International Organizations of Medical Sciences; the International Unions of Biological and of Physiological Sciences; and by direct assistance other than cash grants by Unesco. It consists of: Prof. J. H. Maisin (Belgium), chairman of the Committee and representative of the Council for International Organizations of Medical Sciences; Prof. W. K. Hirschfeld (Holland), vice-chairman; Prof. G. Montalenti (Italy), representing the International Union of Biological Sciences; Prof. J. A. B. Gray (United Kingdom), representing the International Union of Physiological Sciences; three members co-opted under the by-laws for a two-year term from October 1958: namely, Dr. Dale W. Jenkins (United States), representing the International Committee on Laboratory Animals in the American Continent; Monsieur M. Sabourdy (France), treasurer; and Dr. W. Lane-Petter (United Kingdom), secretary-general; and an observer from Unesco. Meetings of this Committee have been held in December 1957 (Paris), October 1958 (Paris) and April 1959 (London). The next meeting will be held in April 1960 in Rome.

Already in December 1956, Unesco had initiated surveys on the production and utilization of laboratory animals in a number of countries, and further surveys have since been commissioned and carried out, on the advice of the Committee. Some have been published, in consolidated form, by the Committee as mimeographed documents: Vol. 1 including India, Italy, Japan, Switzerland and the United Kingdom: Vol. 2, Denmark, Finland, Iceland, Norway, Sweden, and Turkey (first supplement); Federal Republic of Germany and Austria (second supplement); and Australia (third supplement). A third volume will include Belgium, Netherlands, France, Israel, Poland, Czechoslovakia, and probably a partial survey of the United States.

From these surveys has come a clear picture of the actual production and utilization of laboratory animals in many different countries; of the varying special problems, and of the underlying basic needs, which are everywhere the same; and an indication of the way in which an international organization such as the Committee can make itself most useful.

As a further attempt to clarify the task that the Committee had undertaken, a symposium was held in Paris in October 1958; a report of this is still in preparation, but will appear very shortly. At the symposium three main questions were examined. First, what were the detailed requirements in the main biological disciplines using animals: namely, cancer research, radiobiology, microbiology, physiology, cell and tissue culture, endocrinology, and pharmacology and the pharmaceutical industry. Secondly, to what extent were these needs being met to-day in different countries. Thirdly, what were the future possibilities, with special reference to genetics, the control of latent infections, the introduction of new species into laboratory use, and the problems of large-scale production of familiar species.

Since September 1957, the Committee's *Bulletin* has appeared regularly every six months. It contains current news of the work of the Committee and also an excellent bibliographical service on laboratory animals.

Consultant members of the Committee have paid numerous visits to other countries, mainly in Europe, to advise on the creation or organization of national laboratory animal centres and to co-ordinate the efforts being made in this field. Indeed, this sort of personal contact has been of great value in stimulating interest, in encouraging the setting up of new national organizations and in linking the activities of all these in a common effort. In 1956, three national centres only existed, in the United Kingdom, the United States and France. To-day, centres of one kind or another have been formed in Australia, Belgium, the Federal German Republic, Holland, India, Italy, Japan, and more are likely to form in the near future. Almost all owe something, some even their very existence, to the stimulus provided by the Committee.

In 1959, Unesco placed a contract with Monsieur M. Sabourdy, treasurer of the Committee, for the preparation of a list of definitions of terms used in describing laboratory animals of high quality. Such definitions have been attempted in the past, notably among geneticists, but in the absence of a recognized code widely agreed, they tend to lose their exact meaning and in time to become almost valueless. A code of this kind is a necessary adjunct to drawing up specifications for animals; but it should be emphasized that attempts to draw up specifications should not wait on the completion of a glossary of terms, because the need to define many of the terms will become apparent only when specifications are attempted. There must be mutual reaction between the two endeavours, definition and specification.

Most of the surveys already referred to have directed attention to a universal need, namely, for animal technicians who are systematically trained and qualified for the work of breeding and maintaining the animals of high quality that research workers need in nearly every field. Great Britain has been the pioneer in this field, through the work of the Animal Technicians Association, now about ten years old, but training schemes for animal technicians are in operation in United States, France and Czechoslovakia. In the *Bulletin* of September 1959, detailed notes for training animal technicians were published, based largely on the British experience, but taking into account the different needs and opportunities in other countries. The animal technician is coming to take his place alongside the laboratory technician as an indispensable member of the biological team.

The Committee has already given prominence to the need for national laboratory animal organizations, the four functions of which are : (*a*) to act as an exchange for information about all aspects of animal supply and management ; (*b*) to conduct research into current problems in their production and care ; (*c*) to maintain primary type colonies of animals conforming to exacting specifications, supplying authentic breeding stock for expansion in production units ; (*d*) to train scientists and technicians specializing in this field. For the future, it is planning a second symposium for some time in 1961, on transmissible disease (other than of genetic or nutritional origin) ; the preparation of world lists of primary type colonies of specified strains of animals, and sources of less commonly used species of vertebrates and invertebrates ; as well as continuing the labours already undertaken.

The International Committee on Laboratory Animals started as an informal group, largely self-appointed, to examine a problem that was recognized in many quarters, namely the difficulty in obtaining enough laboratory animals of the right quality for current and future research. It is international in the sense that its membership and interests are already widespread, but it is not yet internationally representative. It is still self-recruiting, and no reason so far exists for it to be otherwise, for its present membership has a strong desire to attract the support of all who share its interests.

The very substantial support so far given by Unesco cannot be expected to continue indefinitely, and the grants given by the international unions are not, and probably never can be, sufficient to cover all its actual and intended activities. If the Committee is to carry on as it has started and to develop usefully, other international governmental agencies should be encouraged to help. Of these, the World Health Organization is an obvious candidate for patronage, because of its involvement in medical research, which is so vitally dependent on good laboratory animals. It is satisfactory to note that the World Health Organization has sent an observer to the last two meetings of the Committee.

It would also be reasonable to expect all countries supporting an active laboratory animals organization to have some representation on the Committee. At present only a few enjoy membership of the Committee, and these would appear to have been somewhat arbitrarily selected. There is little doubt that the lop-sidedness of the Committee, in this respect, will be corrected, and the sooner the better.

There is not only a need, but also great scope for international collaboration in defining and producing good laboratory animals. Without such collaboration there is a danger, already unhappily apparent in many parts of the world, of duplicated effort in solving the major problems in this field ; of parallel but isolated developments in the use of animals which appear different but are similar, or appear similar but are differently or misleadingly designated ; and of research effort being vitiated by the use of inadequate animal material. This fragmentation of endeavour has to be corrected before it goes too far. By the end of the decade we have just entered, we shall probably consider, in retrospect, our current ideas of quality in laboratory animals with astonishment bordering on incredulity that they served our purposes even as well as they do. If this is so, the International Committee on Laboratory Animals will have played an important part in our animal house revolution.

TRAINING SKILLED WORKERS

ALTHOUGH much is made of the skill of British craftsmen and operators, investigations over the past decade have shown how little is known about the components of skill. The result has been that much of the training provided for manual workers is conducted in a hit-or-miss fashion because those concerned with the training are not at all clear about the nature of the skill they are trying to impart and, consequently, even less clear about the teaching of it. One whose researches have been almost entirely concerned with the teaching of skills to unskilled and semi-skilled operatives in a variety of industries is W. D. Seymour, and, in a broadsheet published by the Institute of Personnel Management, he presents some of the experience he has gained in training unskilled and semi-skilled operatives in manufacturing industry. This broadsheet should be warmly welcomed by all concerned with the training of operatives as the clearest and most concise statement which has yet appeared.

The types of work customarily undertaken by unskilled and semi-skilled workers in manufacturing industry can be grouped into the following categories :

Handwork, that is, work which is done exclusively with the operators' bare hands, without the intervention of any tools or machinery. This group of operations still employs a large number of operators and includes wrapping and packing operations, some assembly work, pottery decoration and other examples of very fine work. Some types of inspection work also fall into this category.

Handwork with Tools. This category includes all types of manual work in which hand tools are used, but which do not involve the use of mechanical power. In spite of the increasing mechanization of modern industry, very large numbers of workers are still engaged in these operations with hand tools, which may be of a general nature, for example, screwdrivers and pliers, or may be specially designed for the purpose, for example, the wig-maker's hook. Examples of this type of work are provided by most types of assembly work, pottery figure making, automatic telephone wiring, coil fitting on electric motors, etc.

Single Purpose Machine Work. This category covers all operations where a mechanically or electrically driven machine is used for a single specific purpose. The increasing use of special-purpose machines tends to augment the number of workers employed in this category, though the numbers employed on any one type of machine may be small. Examples are provided by hosiery seaming and linking machines, coil winding machines and cigar making machines.

Multi-Purpose Machine Work. The category includes all types of work done on adaptable power-driven machines, which can be used for a variety of purposes. Typical examples in this category are the capstan lathe and the industrial sewing machine.

Group Machine Work. This category includes those types of work where the operator is responsible for the output from a number or battery of machines, which are automatic or semi-automatic in their operation. Such work is sometimes referred to as machine minding, but frequently involves short complex operations which need to be done efficiently if full production is to be achieved. Automatic capstan-lathe work, weaving and spinning provide examples in this category.

Non-Repetitive Work. Operations in this category are frequently even more important than repetitive work and involve a high degree of responsibility; they include machine setting, loom overlooking or tackling and inspection operations and process work.

Techniques of operator training are now being used in a number of industries in Great Britain, and are also being adopted overseas. Underlying these techniques is the recognition that operator skills in industry are very varied and highly developed, and that training of new workers to acquire such skills must be based on an adequate analysis and understanding of the skills. Furthermore, training must be considered as continuing right up to the point where the new recruit attains the experienced workers' standard of output and quality. Where the skills analysis indicates that a task presents special difficulties of movement or of perception, exercises or devices are designed for preliminary training. Again, after further scrutiny of the analysis, the task is divided into sections suitable for training, special attention being given to those parts which involve perceptual difficulties or decision making. Target standards, equivalent to experienced workers' standards of output and quality, are determined for each section of the work. Trainees, normally in pairs and in a training centre adjacent to, but separate from, the shop, practise the preliminary exercises, then practise the sections of the job and proceed to perform the parts together only when each has been mastered to experienced workers' standard of output and quality. When one cycle of the operation has been performed at experienced workers' standard, trainees attempt 2-, 4- or 8-hr runs, passing to a longer period only when targets have been achieved on the preceding shorter period.

By the use of these techniques in a wide variety of industries, such as mechanical engineering, electrical engineering, screw making, pottery, hosiery, garment and footwear manufacture, knitting, spinning and weaving, and other less common industries, it has been found possible to reduce the time required for new entrants to attain the level of performance of experienced workers to between a half and a third of that previously required. Such a reduction in training time, and the higher productivity achieved, lead to economies which usually far exceed the cost of introducing the training scheme. These techniques are also used for the re-training of existing workers whose productivity is below average. The higher output achieved benefits the operators in higher earnings, and the company in lower overhead costs per unit produced.

In the remainder of the broadsheet Seymour describes the methods he has adopted to analyse the nature of skill and some of the findings. Wider use of these findings could do much to increase the quality of skill among British operatives.

This most useful broadsheet may be obtained from the Institute of Personnel Management, 80 Fetter Lane, London, E.C.4, price 7*s.* 6*d.* T. H. HAWKINS

OBSERVED CHARACTERISTICS OF AN ULTRA-HIGH-FREQUENCY SIGNAL TRAVERSING AN AURORAL DISTURBANCE

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AND

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AN experimental study was made to determine the characteristics of ultra-high-frequency signals propagated through auroral-type disturbances. The experiment was performed by illuminating the Moon from a site in the auroral zone and receiving the reflected signals from the Moon at two receiving sites at mid-latitudes.

The transmitter was located at the Geophysical Institute at College, Alaska. The output power was 10 kW. continuous wave at a frequency of 440 Mc./s. The two receiving sites were located at Shirley Bay, Ontario, near Ottawa, and at the Millstone Hill Field Station at Westford, Massachusetts. Each site used an antenna having two orthogonal polarizations. The signals received were studied carefully to determine whether or not there were any differences when the ionosphere was quiet and when the ionosphere was disturbed by auroral activity.

On at least two occasions a strong auroral disturbance was found in the path to the Moon from the transmitting site at College. On these occasions there were two obvious changes in the characteristics of the received signals: (1) there were rapid fluctuations in the polarization angle; and (2) there was an increase in the rate of fading. However, there was no detectable decrease in average power received during the times of these disturbances.

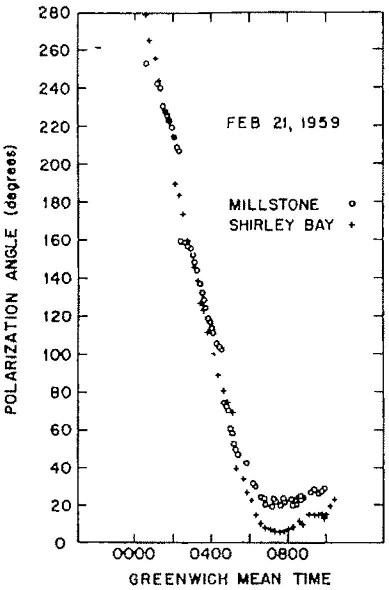

Fig. 1. Variation of the polarization angle of the received signal at the two sites for a day when the ionosphere was quiet

The polarization angle of the signal received was determined by taking the arc tangent of the ratio of voltages in the two orthogonal receiving channels. The quadrant ambiguity resulting from this procedure was resolved by comparing the relative phases of the signals in the two channels before and after a suspected change of quadrant. The polarization angle for a typical lunar day when the ionosphere was quiet is shown in Fig. 1. The polarization angle for this typical run varies slowly with time throughout the day. It was found for these quiet days that the variation of polarization angle could be predicted fairly closely by assuming a certain functional variation of electron density with height, and by adjusting the peak value of this assumed density to match the Central Radio Propagation Laboratory's three-month predictions of 'F-2 Zero MUF'. Account also had to be taken of the relative dipole rotation of the transmitting and receiving antennæ as they tracked the Moon throughout the day with a given polarization.

The measured polarization angles at the two sites for a day when aurora was present are shown in Fig. 2. At about 1400 hr. G.M.T., visual aurora was seen at College in the direction of the Moon. An overcast sky prevented visual observations after about 1500 hr. The rapid fluctuations of polarization angle were apparently caused by the aurora. The computed variation of polarization angle based on a quiet ionosphere is shown by the dashed curve in Fig. 2.

A given change in polarization angle implies a certain change in the number of electrons in the ionosphere, provided certain assumptions are made. One must assume values for the position of the disturbance in electron density and for the magnitude and direction of the magnetic field. On the basis of a stationary magnetic field, the series of fluctuations at 1630 hr. in Fig. 2 represent an incremental change of integrated electron density along a vertical path in the Earth's ionosphere of about 10^{17} electrons per sq. m., assuming that fluctuations in ionization occur below heights of 1,000 km. Very large changes in the direction of the magnetic field are required to explain the observed rapid fluctuations in polarization angle on this basis alone. For example, a change in the direction of the magnetic field of about 20°

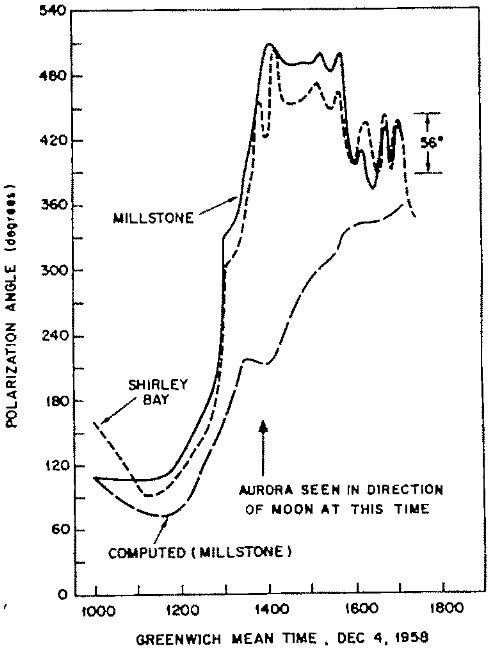

Fig. 2. Variation of the polarization angle of the received signal for a day when the ionosphere was aurorally disturbed. The computed curve is based on a quiet ionosphere

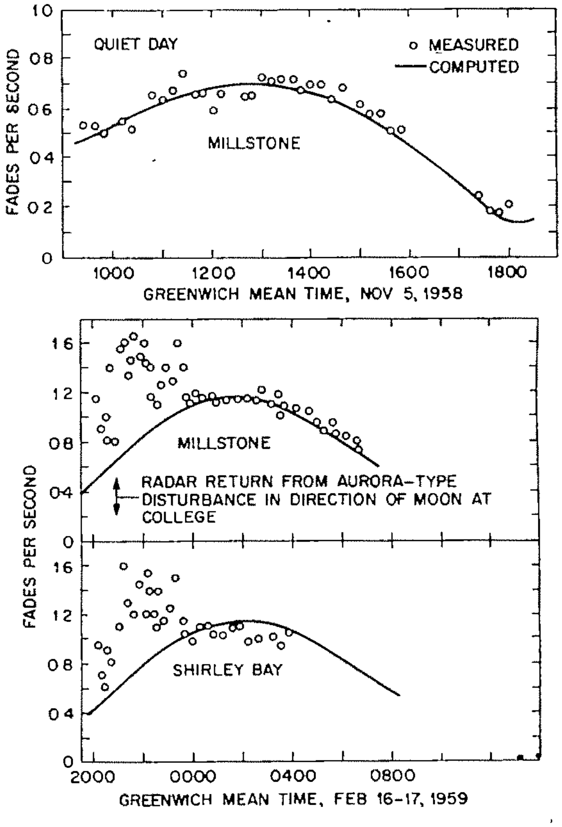

Fig. 3. Comparison of computed and measured rates of fading for a quiet day and an aurorally disturbed day

over a 400-km. height interval in the ionosphere would be required to produce the observed 56° change in polarization angle of Fig. 2 under quiet ionization conditions. This is much larger than any known change in the magnetic field, so most of the rapid fluctuations in polarization angle must be due to rapid changes in ionization density.

During times of an auroral disturbance along the path to the Moon, there was observed an increase in the rate of fading of the received signals. This phenomenon is illustrated in Fig. 3, which shows computed and measured rates of fading for both a quiet day and a disturbed day. During times of a quiet ionosphere, the rate of fading of the received signal was found to be directly proportional to the libration rate of the Moon, an effect that has been reported by others[1]. The predicted rates of fading shown in Fig. 3 were made on the basis of computed rates of libration of the Moon. The top curve illustrates the good agreement obtained between observed and measured rates of fading for normal days. The two lower curves illustrate the increased rates of fading observed during times of auroral disturbances. At approximately 2100 G.M.T. a strong radar return was observed at College from an auroral disturbance in the direction of the Moon. The measured rate of fading at this time was considerably larger than the predicted value made on the basis of Moon libration.

In summary, the significant results reported here are that an auroral disturbance in the path of propagation causes a rapid fluctuation in the polarization angle of the signal received, and an increase in the rate of fading of the signal received, but no measurable absorption of the signal itself. The observed rapid changes in polarization angle are probably due to changes of electron content of the ionosphere of the order of 10^{17} electrons in a vertical column of 1 sq. m. cross-section.

This experiment was a co-operative programme by the following three laboratories: (1) Lincoln Laboratory, a centre for research operated by the Massachusetts Institute of Technology with the joint support of the U.S. Army, Navy and Air Force; (2) Stanford Research Institute, the work of which was sponsored by the Rome Air Development Center of the Air Research and Development Command under Contract *AF* 30 (602)-1762; and (3) the Defence Research Telecommunications Establishment of the Defence Research Board of Canada, the work of which was performed under Project No. *PCC D*48-38-01-11.

We wish to acknowledge the contributions made by our associates at the Lincoln Laboratory, at the Defence Research Telecommunications Establishment, and at the Stanford Research Institute throughout the course of the experiment. We are also grateful for the assistance furnished by personnel of the Geophysical Institute and of the Coast and Geodetic Survey Laboratory at College, Alaska.

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[1] See, for example, Kerr, F. J., and Shain, C. A., *Proc. Inst. Rad. Eng.*, 39, 230 (1951). Browne, I. C., Evans, J. V., Hargreaves, J. K., and Murray, W. A. S., *Proc. Phys. Soc.*, 69*B*, 901 (1956).

OXIDATION OF HYDROXYLAMINE IN CELL-FREE EXTRACTS OF *NITROSOMONAS EUROPAEA*

By DR. D. J. D. NICHOLAS* and DR. O. T. G. JONES*

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WE have prepared cell-free extracts of *Nitrosomonas* that will oxidize hydroxylamine to nitrite provided a suitable acceptor such as cytochrome *c* or phenazine methosulphate is added to the extract or to protein fractions prepared from it. Since this is probably the first report of a cell-free system capable of oxidizing hydroxylamine, we give here a brief summary of our main findings.

Cultures of *Nitrosomonas europaea* (kindly supplied by Dr. Jane Meiklejohn, of Rothamsted Experimental Station) were grown in a culture solution based on that used by Meiklejohn[1] and later modified by Walker (private communication): ($(NH_4)_2SO_4$, 0·66 gm.; NaCl, 0·2 gm.; K_2HPO_4, 0·13 gm.; $MgSO_4.7H_2O$, 0·24 gm.; $FeSO_4.7H_2O$, 0·5 gm. adjusted with 5 per cent w/v K_2CO_3 to *p*H 8·5, glass-distilled water to 1 litre). Phenol red was used as an internal indicator and the *p*H of the medium was maintained constant by titrating it continuously with sterile 10 per cent w/v potassium carbonate. Batch cultures of bacteria (300 l.) were grown at the Medical Research Council Antibiotics Research Station, Clevedon, through the courtesy of the director, Mr. B. K. Kelly, and with the help of Dr. R. W. Harrison; their assistance is gratefully acknowledged.

The bacteria suspended in 0·01 *M* phosphate and 0·05 *M* sodium borate buffer (*p*H 8·3) were disintegrated by means of a Mullard ultrasonic probe for 15 min. at 4° C. in a double-walled glass vessel through which iced water was circulated. The homogenate was centrifuged at 20,000*g* for 20 min. at 4° C.; the supernatant solution was used as the source of the crude enzyme and for subsequent fractionation. Enzyme activity was followed by measuring nitrite production from hydroxylamine in the following reaction mixture: 0·1 ml. enzyme, 0·1 μmole hydroxylamine hydrochloride, 0·2 μmole cytochrome *c* (95 per cent pure, Sigma Chemical Company, U.S.A.) or 0·2 μmole phenazine methosulphate contained in 1 ml. of the phosphate–borate buffer (*p*H 8·3). After the stated incubation period (15 or 20 min.), 0·5 ml. of 1 per cent w/v sulphanilamide in *N* hydrochloric acid and 1 ml. of 0·01 per cent w/v N-(1-naphthyl)ethylenediamine dihydrochloride were added and the volume made to 5 ml. with distilled water, and nitrite determined in a 'Spekker' absorptiometer. Enzyme activity was also determined by following oxygen uptake in a Warburg

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Table 1. EFFECTS OF ANIONS ON NITRITE FORMATION FROM HYDROXYLAMINE IN CELL-FREE EXTRACTS OF *Nitrosomonas europaea* USING CYTOCHROME *c* AS THE ACCEPTOR

	Citrate	Selenate	Arsenate	Tungstate	Phosphate	Pyrophosphate
Per cent inhibition	93	93	85	95	84	90

Silicate and tellurate had no effect.
Reaction mixture: 50 μmoles of the anion, 0·1 μmole cytochrome *c*, 0·1 μmole hydroxylamine hydrochloride in a final volume of 1 ml., phosphate–borate buffer (*p*H 8·8), incubated for 20 min. at 30° C.
Nitrite assayed as in text.

Table 2. FRACTIONATION OF THE ENZYME SYSTEM IN CELL-FREE EXTRACTS OF *Nitrosomonas europaea* THAT OXIDIZES HYDROXYLAMINE TO NITRITE USING PHENAZINE METHOSULPHATE AS THE ACCEPTOR

Fraction	Volume (ml.)	Total units (mμ*M* nitrite/20 min.)	Total protein (mgm.)	Specific activity (mμ*M* nitrite/20 min./mgm. protein)	Purification (fold)	Percentage recovery of enzyme
(1) Crude extract after centrifuging at 20,000*g* for 20 min.	20	6,500	37·4	192		
(2) Fraction collected between 80 and 90 per cent saturation with ammonium sulphate dissolved in 0·01 *M* phosphate and 0 005 *M* borate buffer (*p*H 8·4)	10	2,850	1·1	5,200	27	44
(3) 5 ml. aliquots of (2) put on a *D.E.A.E.*-cellulose column (15 cm. × 1 2 cm.). Column washed in turn with 10-ml. aliquots of the following buffers (*p*H 8·5): 0·05 *M*, 0 1 *M* phosphate-borate; 0·25 *M*, 0 5 *M* phosphate and finally 0·2 *M* pyrophosphate Activity of pyrophosphate eluate	5	510	0 2	8,500	44	8

apparatus in the presence of phenazine methosulphate or cytochrome *c*.

The enzyme activity, which was linear with time when either phenazine methosulphate or cytochrome *c* was used as acceptor, has a *p*H optimum between 7·0 and 8·6. Maximum activity was obtained with 0·2 μmole of either acceptor per 0·1 ml. crude extract containing about 1 mgm. protein per ml. Phenazine methosulphate was the best acceptor, and pyocyanine, methylene blue, benzyl viologen or cytochrome *c* were usually less active, whereas ferricyanide, 2.3.6.-trichloroindophenol dye, coenzymes I and II or glutathione had little effect. Similar results were obtained with whole cells.

When cytochrome *c* was the acceptor, phosphate was required, but at higher concentrations the anion inhibited the oxidation of hydroxylamine as shown in Fig. 1. The phosphate inhibition, which was competitive for cytochrome *c* as shown in Fig. 2, did not occur with phenazine methosulphate. The extent of the inhibition by various anions including pyrophosphate, citrate, tungstate, selenate and arsenate is shown in Table 1. Similar results were

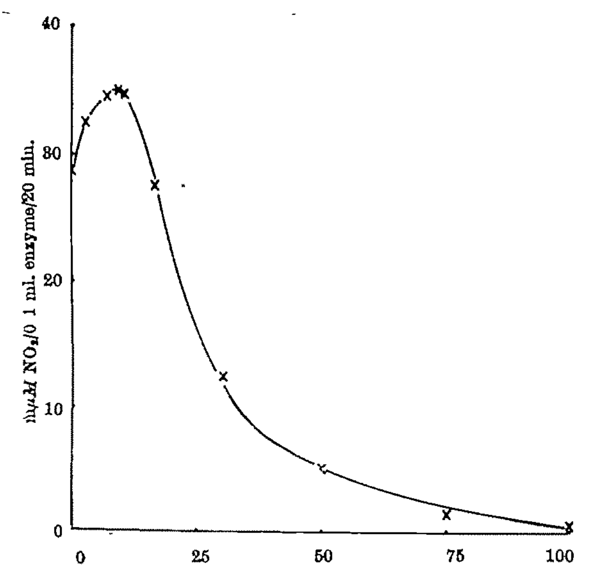

Fig. 1. Effect of phosphate on the oxidation of hydroxylamine to nitrite in a crude extract of *Nitrosomonas europaea*, well dialysed for 12 hr. against 0·01 *M tris* (hydroxymethyl) amino methane buffer, *p*H 8·5
Reaction mixture: 10 μmoles *tris* buffer (*p*H 8 5); 0·2 μmole cytochrome *c*, 0 1 μmole hydroxylamine hydrochloride, 0·1 ml. enzyme contained in 1 ml. incubated for 20 min. at 30° C. Nitrite determined as described in text

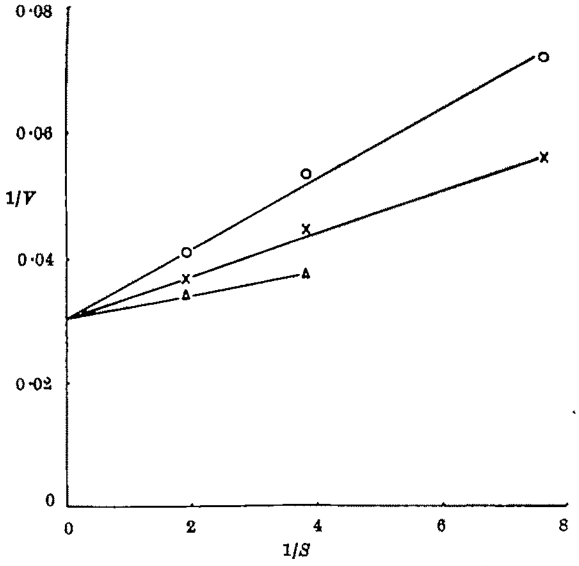

Fig. 2. A Lineweaver–Burk plot for phosphate inhibition of the oxidation of hydroxylamine to nitrite in crude extracts of *Nitrosomonas europaea*, dialysed as described under Fig. 1, using cytochrome *c* as the acceptor
Reaction mixture: 10 μmoles *tris* buffer (*p*H 8·5), 0·1 μmole hydroxylamine hydrochloride, 0·1 ml. enzyme and cytochrome *c* as indicated, contained in 1 ml. incubated for 20 min. at 30° C. Nitrite determined as described in text
1/*S*, reciprocal of cytochrome *c* concentration in mgm. per reaction mixture; 1/*V*, reciprocal of mμ*M* nitrite formed/0 1 ml. enzyme/20 min.

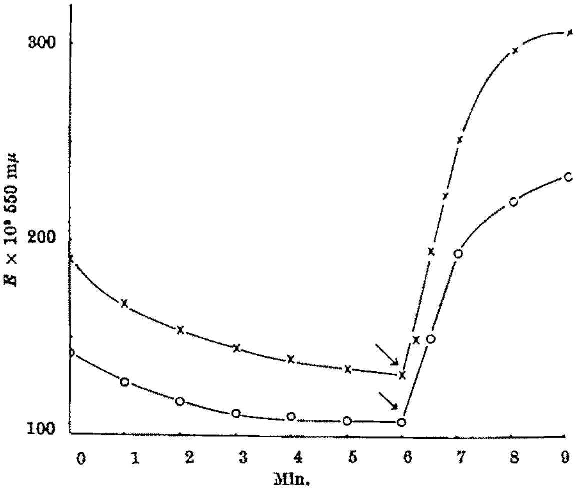

Fig. 3. Effect of hydroxylamine and hydrazine on the enzymic reduction of cytochrome *c* in cell-free extracts of *Nitrosomonas europaea*

+——+, ↓, addition of 0·2 μmole hydroxylamine hydrochloride; ○——○, ↓, addition of 0·2 μmole hydrazine

Reaction mixture in a 1-cm. spectrophotometer cell: 25 μmoles phosphate, 5 μmoles sodium borate buffer (*p*H 8·5), 0·05 μmole cytochrome *c*, in a final volume of 3 ml. and enzymic oxidation of cytochrome *c* allowed to go to completion, then 0·2 μmole of hydroxylamine or hydrazine added

Reaction followed in a Unicam *S.P.*500 spectrophotometer at 550 mμ. Hyponitrite (0·2 μmole), when added instead of hydroxylamine, had no effect. No oxidation or reduction of cytochrome *c* was observed with a boiled enzyme

obtained with whole cells. Hydrazine, a well-known inhibitor of nitrite formation from hydroxylamine in washed cells of *Nitrosomonas*[1], had a similar effect on the cell-free system when cytochrome *c* was the acceptor; but there was no inhibition with phenazine methosulphate. The results in Fig. 3 show that there was an active cytochrome oxidase in the extracts, and the subsequent addition of hydroxylamine resulted in an immediate enzymic reduction of cytochrome *c* with the formation of nitrite. The addition of hydrazine instead of hydroxylamine produced an enzymic reduction of cytochrome *c*, but nitrite was not produced. An increase in hydroxylamine concentration decreased the inhibition of nitrite production by hydrazine. Thus the hydrazine inhibition of nitrite formation in whole cells may be due to a competition with hydroxylamine for a common acceptor such as cytochrome *c* which is indispensable for the oxidation of hydroxylamine. Cyanide inhibited when a range of acceptors was used but carbon monoxide, although depressing cytochrome oxidase activity, did not reduce the oxidation of hydroxylamine to nitrite.

Neither hyponitrite nor ammonia alone was oxidized by these extracts, but the addition of ammonia markedly stimulated the oxidation of hydroxylamine; for example, addition of ammonium sulphate to the reaction mixture at five times the concentration of hydroxylamine stimulated nitrite production by about 60 per cent. Another interesting phenomenon is that the nitrite formed from hydroxylamine in these extracts using cytochrome *c* or phenazine is only 40–73 per cent of the theoretical values expected from the oxygen uptake results obtained in the Warburg. The hydroxylamine was completely oxidized, as confirmed by analysis[3], in agreement with the oxygen uptake. Oxidation with iodine, boiling or acid treatment before the nitrite test did not increase nitrite. When the extracts were aged in the refrigerator for a week or so the nitrite values in the enzyme assays increased, but at best they only approximated to 73 per cent of the theoretical amounts.

A forty-four-fold purification of the enzyme system was achieved by precipitation with ammonium sulphate and further fractionation on diethylaminoethyl-cellulose columns. The pyrophosphate eluate of this column contains the active preparation, using phenazine methosulphate, since pyrophosphate inhibits when cytochrome *c* is the acceptor. The final fraction may contain less protein than that recorded, since pyrophosphate elutes non-protein material from the diethylaminoethyl–cellulose column which reacts with the Folin protein reagents and also absorbs at 260 and 280 mμ.

A more detailed account of this work is in preparation.

[1] Meiklejohn, J., *J. Gen. Microbiol.*, 8, 58 (1953).

[2] Lees, H., "Biochemistry of Autotrophic Bacteria" (Butterworths Publications, London, 1955).

[3] Frear, D. S., and Burrell, R. C., *Anal. Chem.*, 27, 1664 (1955).

RESPONSE OF INBRED AND F_1 HYBRID MICE TO HORMONE

By DR C. K. CHAI

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IN the hands of the biologists the house-mouse is one of the mammals subjected for a long time to continuous mating between close relatives, thereby having attained a high degree of inbreeding and the greatest number of inbred lines with distinct biological characteristics. Because of its biological and genetic uniqueness, and its high rate of reproduction and ease of maintenance, this small-sized mammal has gained recognition as an experimental animal of great value in genetic and medical research and in biological assay. It is on the use of the mouse in bioassays that questions have been raised as to whether inbred mice are superior to random-bred mice and to animals of equal genetic homogeneity but heterozygous, such as F_1 hybrids, produced by crossing two inbred strains.

About two decades ago Emmens[1] found that some F_1 hybrid mice were less variable in response to œstrogen than their inbred parental strains. Later, Mather[2] presented some genetic considerations involved in biological responses. Recently, some data have been presented by McLaren and Michie[3], Claringbold and Biggers[4], and McLaren and Michie[5], and a brief review has been made by Biggers, McLaren and Michie[6] concerning the suitability of using inbred animals as compared with F_1 hybrid animals and random-bred stocks in biological assays. It has been claimed by the latter that inbred animals are not desirable for bioassay on the grounds that they are more variable in responses than either the F_1 hybrid or random-bred animals.

The problem concerning the choice of assay animals requires further consideration. Biological responses involve the reactions in the animal body of one to many systems, all of which are controlled to different extents by the genetic constitution of the individual. The nature of the responses is complicated, and the end-points used are different from one assay to another. However, in determining the precision of an assay, an important parameter which must be considered in quantitative assays—in addition to variance—is sensitivity of response as measured by the slope of the dose–response curve.

This article, which presents some of my experimental results and reviews briefly the results published by others, concerns mainly the sensitivity of response to hormonal substances based on the principles of bioassay and experimental evidence from mice. Some genetic interpretations and general conclusions are made. Although the evidence is drawn from experiments with hormones as assay substances and mice as assay animals, the conclusions reached may be applicable to other drugs and biological preparations and other animal species.

Requirements of an Assay Animal

Biological assays may be defined as determinations of potency or toxicity of a given substance based upon the response of living matter. The nature of the response may be : (1) alteration of internal state —adjustment ; or (2) maintenance of relative constancy of the internal state—regulation. It is very likely that most biological stimuli cause both mechanisms to operate at the same time. For convenience of measurement, biological responses are ordinarily classified into two main categories : (1) qualitative or all-or-none response ; and (2) quantitative. A qualitative response is measured by the percentage of assay subjects with a positive reaction. The most common qualitative data are quantal. An alternative method for quantal data is the use of reaction time, which is relatively seldom used in hormone assays. A quantitative response is very commonly used in hormone assays ; it should give more information per observation than a quantal reaction.

The design of the assay and its computation depend mainly upon the characteristics of the response to be considered. The basic principles for satisfactory assay by Gaddum[7] may be adapted as a guide in the choice of an assay animal. These principles are : (1) a linear relationship should exist between response and dose over the widest possible range of dose ; (2) the standard deviation should be independent of the response ; (3) a minimal value for the ratio s^2/b^2 in quantitative assay and $1/b^2$ in quantal assay, where s^2 is the within-group variance and b is the slope of the dose-response line. (This ratio is called λ^2 by Gaddum.) In addition, good assay animals should give reproducible results, and the precision of an assay should not alter much from test to test or from time to time.

The answer to the question as to whether F_1 hybrids are preferable to inbreds in hormone assay can be derived from calculations based on the formula $\lambda = s/b$ in quantitative assay and $\lambda = 1/b$ in quantal assay. Inbred strains, or F_1 hybrids, which give the smallest λ, offer the most precise assay information or the most information per animal.

In quantitative response, comparison of the assay value between the inbreds and F_1 hybrid animals is therefore based on the ratio of s to b. The ratio may not be minimal when s is minimal, since it depends on the relationship between the two quantities. A slope which is steep enough may compensate for a large variance. Thus, the choice of a mouse strain or F_1 hybrid for bioassay of a given substance is governed by the two parameters in quantitative assay, rather than by either one alone.

In assays involving the quantal response, the precision of the assays depends entirely on b. The value of b, in fact, measures the variation in response of the particular group of animals, for the variances are entirely dependent on b, since $s^2 = 1/b^2$. Unless reliable information is available, the choice of optimum dose-level and adequacy of transformation is frequently a troublesome matter in practice, especially in comparisons of genetically different groups of animals. Inadequate transformations or unsuitable dose-levels easily lead to biased estimates of the true value of b, which in turn results in biased estimations of variance. For this reason the quantitative responses are considered to be more appropriate than the quantal responses in comparing the bioassay value between genetically different groups of animals.

Difference in Sensitivity or Magnitude of Response

Sensitivity is measured by the slope of the dose–response curve. Magnitude of response at a given dose may also be indicative of sensitivity but only when comparing dose–response curves which (1) originate from approximately the same point, and (2) are linear after appropriate transformation. The following examples cover mice of many inbred strains and F_1 hybrids popularly used in bioassays of various hormones. Some of my unpublished results are given first and followed with a tabulation of results published from this and other laboratories.

Response to Androgen. Mice of the *C57BL/6* and *C57BR/cd* inbred strains and their F_1 hybrid BBF_1 were used in this experiment. (The two inbred strains were comparatively closely related and their biological characteristics are rather similar.) The mice were weaned at about four weeks of age, and only those of 9–12 gm. body-weight were used. They were castrated within three days after weaning. Animals were divided into dose groups (eight animals each) in each strain and in the F_1 hybrid mice, and each dose group received a different dose of injection of testosterone in liquid suspension. The injection was begun on the day following castration, one dose a day, and continued for twenty days. The mice were weighed and autopsied on the day following the last injection. The seminal vesicles were removed, blotted thoroughly, and weighed. The means and variances of seminal vesicle weights in each dose group are given in Table 1 and illustrated in Fig. 1. (The methods were similar to a previous experiment using testosterone propionate in different inbred strains of mice—for details see Chai[8].) There

Table 1. MEANS AND VARIANCES OF SEMINAL VESICLE WEIGHTS IN RESPONSE TO TESTOSTERONE IN INBRED AND F_1 HYBRID MICE

Dose (mgm.)	*C57BL/6* mean (mgm.)	s^2	*C57BR/cd* mean	s^2	F_1 (*C57BL/6* × *C57BR/cd*) mean	s^2
0·016	1 9	0 19	2 9	1·33	1·5	0 15
0·032	2·3	1 09	4·4	1 33	3·4	0 58
0·064	6·6	1·10	12 5	4·41	4·7	1·71
0·128	20·3	6·39	24·4	7 25	19·7	16 79
0·256	26·9	35 63	28·8	19·10	29·8	4·97

Table 2. REGRESSION OF RESPONSE IN PERCENTAGE OF OVARIAN FOLLICLES TRANSFORMED TO PROBITS ON SERIAL DOSES OF HCG IN INBRED AND F_1 HYBRID MICE

Genetic groups	No. mice	Regression equations* ($a + bx$)
DBA/1	80	$4{\cdot}91 + 1{\cdot}25x$
DBA/2	80	$4{\cdot}96 + 0{\cdot}99x$
A	88	$4{\cdot}69 + 0\ 62x$
CAF_1	80	$5{\cdot}19 + 0\ 64x$
BALB/c	70	$5\ 61 + 1{\cdot}27x$

* a, fitted constant; b, slope; x, log dose.

appears to be no difference in the slope of the dose–response curves between the three different groups of mice. The F_1 hybrid was no more sensitive than either one of the parental strains in this particular response.

Response to Gonadotrophin. (The following forms a part of the results of an experiment carried out by J. Leopold, as a summer student under my guidance.) Female mice about one month of age from inbred strains, *DBA/1*, *DBA/2*, *BALB/c*, *A* and the F_1 hybrid of the last-named two strains, were divided into dose groups. Chorionic gonadotrophin was serially diluted. One dose was assigned to each group and two injections were given to each mouse at 24-hr. intervals. Autopsy was made at about 12 hr. after the last injection. The presence of hæmorrhagic follicles in the ovaries was considered a positive response. The percentage of animals giving a positive response was calculated for each dose group. Probit transformation was made, and a linear regression was computed for each strain and hybrid according to the methods described by Finney[9]. The results are given in Table 2. The regression of the dose response appears to be largest for the *BALB/c* and least for the *A* strain; that of the F_1 hybrid fell between those of the parental mouse strains but close to that of the *A* strain. The difference in slope between the two *DBA* sublines, though not large, seems to show their genetic differences in the response as a consequence of separation in breeding for about twenty generations.

The published results using various hormones as sources of stimulation are given in Table 3. In addition to the differences in responses shown among inbred strains, out of nineteen comparisons of responses between F_1 hybrid mice and their parental inbred strains, ten hybrids fell between their parental strains, four were comparable to, and five greater than, either one of the parents.

Genetic Basis of Hormonal Response

A biological response, either physiological or morphological, varies within limits set by the genotype, although it is influenced greatly by environment. This is clearly indicated by: (1) the differences in response to various hormones between the inbred strains in the given examples; (2) response to selection for sensitivity to gonadotrophin[22]; (3) relative proportion of genetic to environmental variance in gonadotrophin assay[23]; (4) the heritability of iodine metabolism in the thyroids of mice[24], and the difference in response to thyroxine between inbred strains

Table 3. RELATIVE SENSITIVITY OR MAGNITUDE OF RESPONSE TO HORMONES IN INBRED AND F_1 HYBRID MICE

Hormone applied	Criterion of response	Relative sensitivity* or magnitude of response	Author
Œstrogen	Vaginal smear	$C_{57} > C_{57} \times CBA > CBA$ $C_{57} > CBA \times C_{57} > CBA$ $CBA \sim CBA \times A \sim A$	Trentin (ref. 10)
Œstrogen	Vaginal smear	$C_{57}BL > C3H \sim C$	Shimpkin (ref. 11)
Œstrone Intravaginal	Vaginal smear	$C_{57} \times CBA \sim CBA \times C_{57} > CBA > C_{57}$	Claringbold and Biggers (ref. 4)
Œstrone-3 : 17*B* Subcutaneous		$C_{57} \times CBA \nless CBA \times C_{57} > CBA$	
Œstrone and pregesterone	Mammary gland area	Castrate male $C3H > A > BALB/c > C57BL$ $BALB/c > BALB/c \times A > A$ $BALB/c > A \times BALB/c$ $C3H > C3H \times C57BL > C57BL \times C3H > C57BL$	Blair (ref. 12)
Mammary-gland stimulating hormone	Mammary lobule alveolar growth	Schwing $\ngtr$ Swiss > Sutter ~ Kansas	Mixner and Turner (ref. 13)
Œstrogen	Adrenal weight (per cent of control)	Both ♂ and ♀ $BALB/c > BALB/c \times C3H > C3H/He$ $BALB/c > BALB/c \times A > A/He$ $C_{57}BL > C57BL \times C3H > C3H$ $C_{57}BL \sim C3H \times C_{57}BL > C3H$	Westberg (ref. 14)
Testosterone propionate	Kidney *B*-glucuronides	$A/Cl > AKR > BAF_1 > C3H > CAF_1 > DBA > 129$	Fishman and Farmelant (ref. 15)
Cortisone	Number of eosinophils	$129 \times BR/cd > C57BL \times C57BR/cd > C57BL/6 > C57BR/cd > ST > C57BR/a > 129/Rr > DBA > BALB/c$	Wragg and Speirs (ref. 16)
Chorionic hormone	?	$C57 > A$	Carr (ref. 17)
Œstrone	Mammary growth Œstrus symptoms	$C57Bl \sim O_{20} > DBA$ $C57BL \sim O_{20}$ = Riet > DBA	Muhlbock (ref. 18)
Œstrone	Mammary growth Per cent vaginal œstrus	(Castrate male) $DBA > C57BL \sim O_{20}$ > Riet $C57BL \sim O_{20} > DBA$	Muhlbock (ref. 19)
Human pregnancy urine	Per cent ovarian follicles	$C57BL \sim C57L > A$	Hummel (ref. 20)
Testosterone propionate	Seminal vesicle weight	$BALB/c > CAF_1 > A/Jax$	Chai (ref. 8)
PTU and thyroxine	Thyroid weight	$A/Jax > BAF_1 > C57BL/6$ $DBA/1 \sim DBA/2 \sim BDF_1 > C57BL/6$	Chai (ref. 21)
" "	Thyroid follicular cell height	$BAF_1 > C57BL/6 \sim A/Jax$ $BDF_1 > DBA/1 \ngtr DBA/2 \ngtr C57BL/6$	

Remarks: $BAF_1 = C57BL/6 \times A/Jax$; $CAF_1 = BALB/c \times A/Jax$; $BDF_1 = C57BL/6 \times DBA/2$

* The sequence is based mainly on observations of graphic illustrations and/or the mean values of responses. The differences between strains and F_1 hybrids in a response as given may not be statistically significant and the original authors might not fully agree with the given order.

of mice and their F_1 hybrids as determined by iodine turnover rate in the thyroids[25].

It may be pointed out particularly that on the results concerning the responses of *BALB/c* and *A* strains and their F_1 hybrids (Table 3), one is the response to œstrone and progesterone, measured by mammary gland area[13], and the other is response to œstrogen as measured by adrenal weight[14]. In both experiments, the F_1 hybrid fell between the parental strains and the order is the same as in assays using androgen[7] and gonadotrophin for stimulation. These results are from different laboratories, but in spite of possible differences in environmental conditions and in assay techniques, the same order of relative sensitivity has been obtained in mouse stocks. This may indicate that the same, or closely linked, genes control some of the related endocrine systems, and contributes further evidence toward the genetic determination of hormonal responses.

Except in a few cases (such as resistance to insulin in obese mice and responsiveness to pituitary extract in dwarf mice) where the action of a single gene can have a significant effect upon certain responses, the inheritance units determining hormonal responses in the mice commonly used in bioassays must be assumed at present to be polygenes. The biological properties of these polygenes may be considered to be the same as those which determine characters generally used in quantitative genetic studies. That is, their effects are small, additive and supplementary. The additive effects of polygenes in general have been found to be more important than dominance and epistatic effects, if the latter exist.

On this basis, the consequence in hormonal responses of F_1 hybrids in comparison to their parental strains may be tentatively postulated. When the parental strains differ greatly in a given biological response, it may be assumed that one strain possesses most of the alleles with plus effect and the other parent alleles with minus effects in the loci concerned. The consequence of combining one from each pair of alleles of these two sets as in the F_1 hybrids would result in an intermediate response or one close to it. It is supposed that in general the dominant effects are weaker than additive effects in polygenic inheritance. Therefore, even the net effects of dominance may not be exactly cancelled but the response in the F_1 will be unlikely to exceed those of either one or the other parental strain in the response concerned. This supposition seems to hold in many animal experiments dealing with quantitative characters and may hold in the present case. On the other hand, when the parental strains are similar in response, it is possible for the F_1 hybrids sometimes to give responses either greater or smaller than either parental strain, as a result of the supplementary effects or interaction of genes with the other genetic background. The responses to testosterone in four inbred strains of mice and two F_1 hybrids (Figs. 1 and 2), and results published by various investigators as given in Table 3, seem, in general, to support the above hypothesis. It is realized that this is a great over-simplification in this brief postulation of the genetic basis of hormonal responses, but, regrettably, until better understanding of the genetic and physiological properties of polygenes are arrived at, further speculation would be impractical.

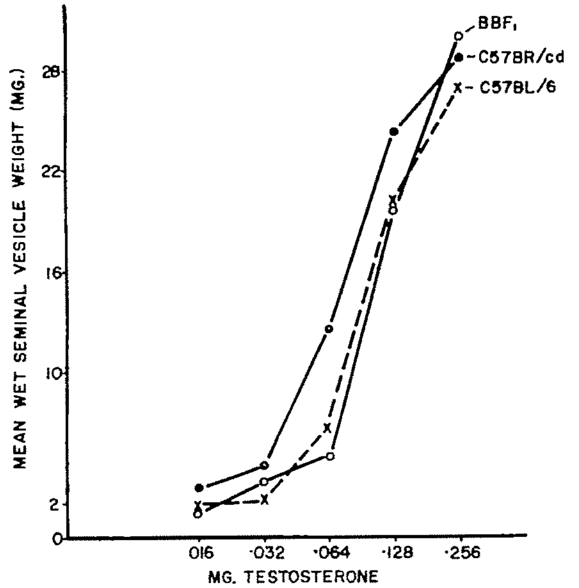

Fig. 1. Showing the response to graded doses of testosterone as measured by wet seminal vesicle weights in *C57BL/6*, *C57BR/cd* and their F_1 hybrid mice, *BBF*$_1$. It appears that there are no differences between them in sensitivity

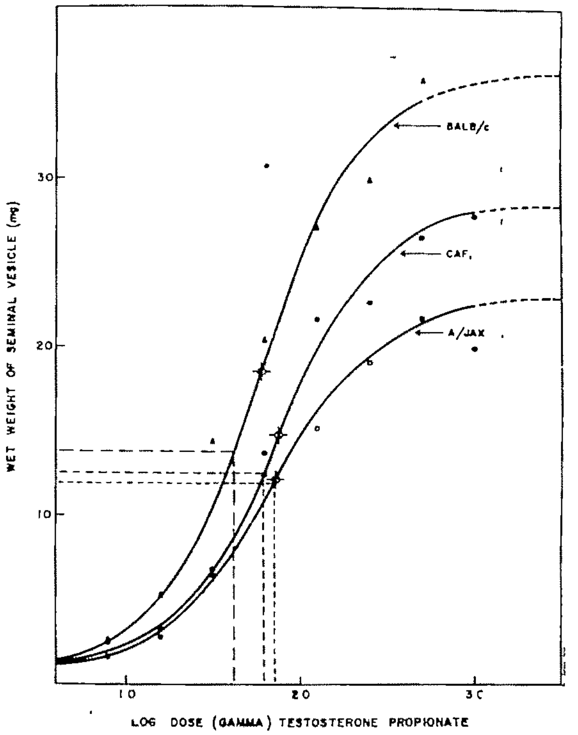

Fig. 2. Showing the distribution of seminal vesicle weights and fitted logistic curves for the *A/Jax*, *BALB/c* strains of mice and their F_1 hybrids treated with graded doses of testosterone propionate. The slope of the dose-response curve of *BALB/c* mice is greater than that of *A/Jax*, and that of their F_1 hybrid is about intermediate between those of the parental strains (from Chai, ref. 8, courtesy of *Anat. Rec.*)

Conclusions

The above data show that in the majority of cases the responses of the F_1 hybrids to various hormone stimulations fell between those of their parental strains. This indicates that the F_1 hybrids are not, in general, preferable to inbred mice in hormone assay in so far as the *b* value is concerned, for *b* is one of the two parameters in quantitative response and

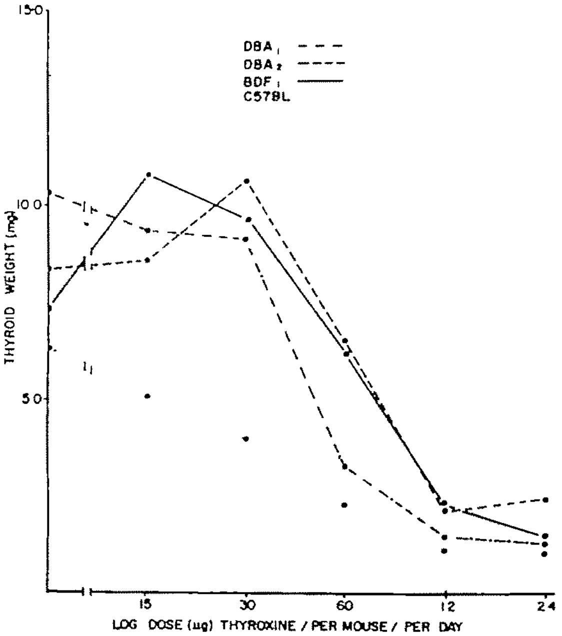

Fig. 3. Distribution of mean thyroid weights in the mice of *C57BL/6*, *DBA/1*, *DBA/2* and *BAF*$_1$ (*C57BL/6* × *DBA/2*). Each point represents a mean thyroid weight at a given dose from eight mice. Each mouse received a subcutaneous injection of thyroxine, one dose a day, for twenty days. All mice were on a diet of Purina Mouse Laboratory Chow mixed with 0 2 per cent of propylthiouracil during the treatment. Note the long linear portion of the curve for *C57BL* mice, which is desirable in bioassay, although the slope of the curve is smaller than that of the curves for the *DBA*'s and the *BAF*$_1$ (from Chai, ref. 21; courtesy of *J. Heredity*)

the only parameter in qualitative response in determining the precision of the assay.

The linear relationship between response and dose over the widest possible range of dose is another matter worthy of consideration. It is noticed that the response to both goitrogen and thyroxine in the *C57BL* mice was distributed linearly throughout practically all the dosages applied, whereas the linear portion for the F_1 hybrids fell within a limited range of dose[21] (Fig. 3). Whether this is due to better self-regulating properties of the F_1 hybrids, such that they can use the hormones more efficiently, is a problem remaining for further exploration.

It is generally claimed that F_1 hybrids are more uniform in growth and development than inbreds and this is, no doubt, a desirable feature in bioassay. The importance of body-size differences affecting the response depends on the degree of relationship between the criteria of response and body-size. In most cases, this can be handled satisfactorily by covariance analysis. The part of the variation in response as caused by differences in body-size should not be interpreted as the intrinsic difference in magnitude of reactivity of the assay animals themselves, but represents nothing more than the phenotypic heterogeneity of the assay subjects. More extensive data are needed to prove differences between the two types of genetically homogeneous populations, allowing for body-size differences. That is, whether there are basic differences in variation of reactivity to stimulation, which have not resulted from pre-existing anatomical or physiological differences in variability, remains to be studied.

It has been suggested that under unfavourable environmental conditions the homozygotes are more variable than the heterozygotes in growth and development. This might apply to biological responses and explain the differences obtained by investigators from different laboratories and the so-called transient effect in some laboratories. It is possible that the F_1 hybrids may stand long-distance shipment and change of environment better than inbreds, due to their better self-regulating ability and resistance to disease. This is a matter which needs to be considered in practice. On the same argument, it may be true that the homozygotes, such as the inbred mice, can be more subject to internal environmental changes of physiology and thus show more sensitive responses in bioassay than do heterozygotes. The recent report by Becker *et al.*[26] comparing growth response of inbred and non-inbred chickens at different nutritional levels seems to support this hypothesis.

In summary, F_1 hybrids do not necessarily have greater sensitivity as measured by the magnitude of responses at each dose-level. That F_1 hybrids are, in general, more uniform in growth and development and better adjusted to environmental changes than inbreds is often claimed. This is expected to affect the variability of response and may be an advantage of F_1 hybrids over inbreds in bioassays. Depending on the genetic background, some inbreds or F_1 hybrids may be more suitable for certain hormone assays than for others, and animals of some genotypes may be less variable in one environment than in another. For the assay of a given substance, the choice of assay animal—either an inbred or an F_1 hybrid—cannot be made in advance; it has to be based either on existing data or determined experimentally.

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[1] Emmens, C. W., *J. Endocrinol.*, **1**, 373 (1939).
[2] Mather, K., *Analyst*, **71**, 407 (1946).
[3] McLaren, A., and Michie, D., *Nature*, **173**, 686 (1954).
[4] Claringbold, P. J., *et al.*, *J. Endocrinol.*, **12**, 9 (1955).
[5] McLaren, A., and Michie, D., *J. Genetics*, **54**, 440 (1956)
[6] Biggers, J. D., McLaren, Anne, and Michie, D., *Nature*, **182**, 77 (1958)
[7] Gaddum, J. H., *Biochem. J.*, **25**, 1, 118 (1931).
[8] Chai, C. K., *Anat. Rec.*, **126**, 269 (1956).
[9] Finney, D. J., "Probit Analysis" (Cambridge Univ. Press, 1952).
[10] Trentin, J. J., *Cancer Res.*, **10**, 580 (1950).
[11] Shimpkin, M. B., and Andervont, H. B., *J. Nat. Cancer Inst.*, **1**, 599 (1941).
[12] Blair, S. M., *et al.*, *Endocrinol*, **61**, 643 (1957).
[13] Mixner, J. P., and Turner, C. W., *Proc. Soc. Exp. Biol. Med.*, **95**, 87 (1957).
[14] Westberg, Jim A., *et al.*, *Acta Endocrin.*, **25**, 70 (1957).
[15] Fishman, W. H., and Farmelant, M. H., *Endocrinol.*, **52**, 536 (1953).
[16] Wiagg, L. E., and Speirs, R. S., *Proc. Soc. Exp. Biol. Med.*, **80**, 680 (1952).
[17] Carr, J. G., *Abst. Heredity*, **3**, 262 (1949).
[18] Muhlbock, O., *Acta Brevia Neerl.*, **15**, No. 1, 18 (1947)
[19] Muhlbock, O., *Acta Brevia Neerl.*, **16**, 1 (1948).
[20] Hummel, Katharine P., *Endocrinol.*, **30**, 74 (1942)
[21] Chai, C. K., *J. Hered.*, **49**, 143 (1958).
[22] Casida, L. E., *et al.*, *Endocrinol.*, **51**, 148 (1952).
[23] Rollins, W. C., and Cole, H. H., *Endocrinol.*, **51**, 203 (1952).
[24] Chai, C. K., Symposium on Biometrical Genetics, held 1958, ed. O. Kempthorne (in the press).
[25] Amin, A., Chai, C. K., and Reineke, E. P., *Amer. J. Physiol.*, **191**, 34 (1957).
[26] Becker, W. A., and Berg, L. P., *Poultry Sci.*, **38**, 362 (1959).

LETTERS TO THE EDITORS

GEO- and RADIO-PHYSICS

A Day-time Maximum of Oblique Auroral Reflexions observed in the Auroral Zone

ALMOST all investigations of high-frequency and very high-frequency reflexions from aurora made in or near the auroral zone have given diurnal variation curves with one main maximum near local midnight[1-3]. An exception is the recent observations by Presnell *et al.*[4] at 216 Mc./s., made at College, which—for diffuse echoes only—has given two maxima, one in the morning and one in the afternoon with the peak between 1700 and 1900 L.T. Observers at lower latitudes have usually found two maxima in the night[5-8].

From four months records (March–June 1959) of aurorally propagated very high-frequency signals taken in the auroral zone at Kiruna Geophysical Observatory (geographic co-ordinates 67·8° N., 20·4° E., geomagnetic co-ordinates 65·3° N., 115·5° E.) and transmitted by the Finnish broadcasting station at Kemi (geographic co-ordinates 65·8° N., 24·8° E., geomagnetic co-ordinates 62·5° N., 117·6° E.) a diurnal variation curve with two maxima has been found (cf. Fig. 1). One of these maxima is in day-time between 1200 and 1600 M.E.T. The experimental points include all observed reflexions. This is in remarkable contrast to the diurnal variation mentioned above, which was observed by radar methods at Kiruna (in 1951–52) and at other places in auroral zone latitudes.

The transmissions studied at Kiruna have a frequency of 92·8 Mc./s. The broadcasting station, 310 km. distant, has sent out continuous waves in the periods between programme transmissions. The radiated power is 8·9 kW. and the polarization horizontal. A 'Servo' model *R* 5200-*A*2 receiver and a five-element Yagi antenna are used. The sensitivity is better than 1 μV. for 50 mW. output for a band-width of 25 kc./s.

There is no doubt that this auroral echo day-time maximum is real. The day-time echoes have the same signal characteristics observable[2] as night echoes[1] on the Esterline Angus records. The direction distributions for day- and night-maxima are approximately the same ; this has been determined by means of rotary antennæ. The day-time maximum has been found also in monitoring another transmitter (Östersund in central Sweden, 87·9 Mc./s.).

The local time for the afternoon echo maximum of Fig. 1 is much earlier than that found by any other observers, so far as we are aware. If the geomagnetic time for the maximum is investigated, the difference from the time of maximum found on the western hemisphere (especially in Alaska) is reduced by several hours, but also the geomagnetic time for the afternoon maximum found at Kiruna is earlier than reported by anybody else. Recent work at a frequency of 216 Mc./s. (ref. 4) has given an afternoon maximum, but only for a special type of echo, the so-called diffuse echoes. The difference between the geomagnetic time for the afternoon maximum reported by Presnell *et al.*[4] and that shown in Fig. 1 is about 1 hr.

The investigation at Kiruna will be extended over a longer period of time, and it is hoped that the recorded material will show if the diurnal variation curve described above is characteristic of observations of oblique auroral very high-frequency reflexions at Kiruna for longer periods, or only for the four months analysed.

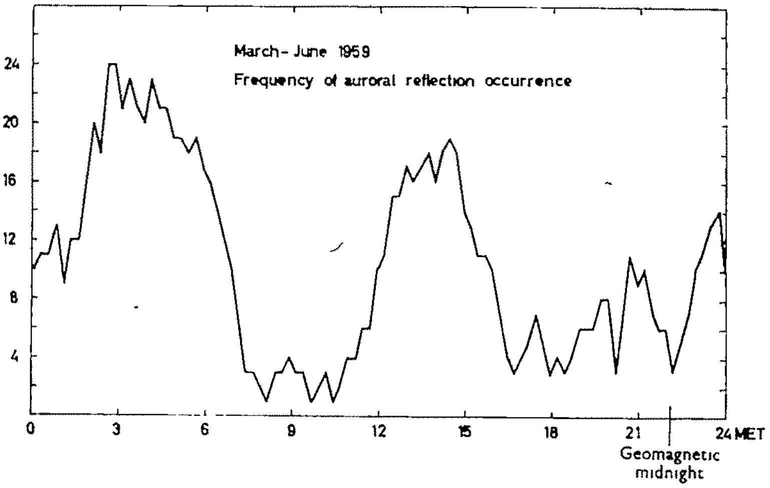

Fig 1

The work on which this communication is based has been supported by the European Office of Air Research and Development Command (Contract No. *AF* 61(514)-1314).

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[1] Hellgren, G , and Meos, J , *Tellus*, **3**, 249 (1952)

[2] Bowles, K. L., Dyce, R. B., and Little, C G., Final Report Task E. Signal Corps Contract No. *DA*-36-039-*SC*-56739, Geophysical Institute, College, Alaska (1955).

[3] Currie, B. W., Forsyth, P A., and Vawter, F. E., *J. Geophys. Res.*, **58**, 179 (1953).

[4] Presnell, R. I., Leadabrand, R. L., Peterson, A. M., Dyce, R. B., Schlobohm, J C., and Berg, M R , *J Geophys Res.*, **64**, 1179 (1959)

[5] Boullough, K., and Kaiser, T. R., *J. Atm. Terr. Phys.*, **6**, 198 (1955).

[6] Dyce, R , *Trans Inst. Rad. Eng.*, *PGAP*-3, **2**, 76 (1955).

[7] Gerson, N. C., *Proc. Phys. Soc* , **68**, B, 408 (1955).

[8] Unwin, R. S., *Ann Géophys.*, **15**, 377 (1959).

Observations of Ionization induced by Artificial Earth Satellites

While monitoring a remote continuous-wave station at the Ohio State University Radio Observatory, it has been found that large enhancements of signals often occur at the times of near approach of an artificial Earth satellite. The enhancements are of such magnitude as to indicate extensive ionization with scattering cross-section several orders of magnitude greater than the physical cross-section of the satellite[1-3]. In these observations it has been convenient to monitor the National Bureau of Standards time service station *WWV* near Washington, D.C., at a distance of 330 miles from Columbus. This communication describes some additional observations of the phenomenon.

Between January 9 and February 7, 1959, the enhancements of signals of *WWV* on 20 Mc./s. were recorded on three or four passes of *Sputnik III* each night (between local midnight and 6 a.m.), while recordings of the satellite transmissions on 20·0045 Mc./s. were recorded to determine the time of near approach. The passes during this period were typically west to east with near approach over Hudson Bay (north of Columbus). A three-element beam antenna directed to the north was used for reception. The *WWV* records were divided into approximately 100-min. intervals centred on the near approach time of *Sputnik III*. All *WWV* enhancements of more than a constant minimum magnitude and more than 30 sec. in duration were taken as significant. Adding all such enhancements (about 200) for a total of 50 passes on 14 nights (3–4 passes per night) a histogram was obtained. Averages for each 5-min. period were taken and a smoothed curve drawn through these values. The result is shown in Fig. 1.

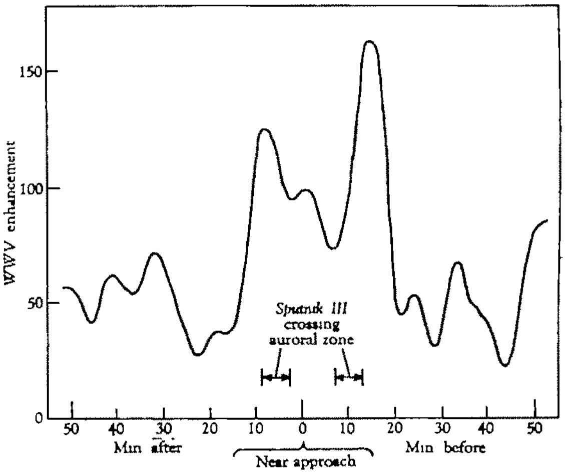

Fig 1. Enhancements of *WWV* 20-Mc./s. signals as a function of time before and after the near approach of *Sputnik III* for 50 passes on 14 nights during January and February 1959

This graph gives the amount of *WWV* enhancement (number of enhancements times magnitude) to an arbitrary scale as a function of time before and after the near approach of *Sputnik III*. A peak occurs at the time of near approach, but larger peaks somewhat before and after. These three peaks, grouped around the near approach, are as much as 10 times greater than the standard deviation in the remainder of the graph, which makes it very unlikely that they are due to a random process. It is to be noted that the largest peaks are 15 min. before (precursor enhancement) and 8 min. after near approach (post-enhancement). It may be significant that these times correspond approximately to the times the satellite was, on the average, passing through the auroral zone (see Fig. 1). A possible hypothesis for explaining this effect is that the satellite encountered regions of high-particle density in the auroral zone, scattering material into ionized clouds or streams which travelled closer to Columbus with reflexions from these clouds causing many of the observed signal enhancements.

In order to obtain more data on the ionized clouds, a radar transmitter of about 600-W. peak power was installed at the prime focus of the Ohio State University 360-ft. radio telescope[4]. At the time, the parabola was completed but the flat reflector had not been assembled, so that the parabola was unobstructed to the north. Using frequencies between 17 and 19 Mc./s., a fan beam about 10° wide by 50° high was produced above the northern horizon, with a gain of nearly 20 decibels. The receiver was located about 20 miles south at Columbus. The receiver output was displayed on a *z*-type oscilloscope and was recorded on 35-mm. film moving 2 cm./min. With this arrangement a number of significant records were obtained. Fig. 2 shows one of these, obtained on February 20, 1959. Time increases to the left and range increases vertically downward. Between 11.13 and 11.15 p.m., *Sputnik III* was in the beam of the 360-ft. parabola travelling north-west to south-east at an average range of about 1,400 miles (height 860 miles). At 11.13 several traces appear on the film which radiate from the point *A*. At 11.13·8 two traces appear on the film which radiate from the point *B* and at 11.14·8 there are two traces radiating from point *C*. Points *A*, *B* and *C* are all close to the range of the satellite (dashed line) as calculated from ephemeris and other data with the satellite reaching its point of near approach north-east of Columbus about 11.17 p.m. at a range of about 1,200 miles. At point *C* there are two faint traces close to the range of the satellite. These may be due to reflexions from ionization at or close to the satellite.

It is postulated that at points *A*, *B* and *C* the satellite encountered regions of high density scattering a number of particle clouds. Clouds from the radiant points *B* and *C* approached within about 400 miles of Columbus. (Similar particle clouds may also account for the precursor enhancement peak in

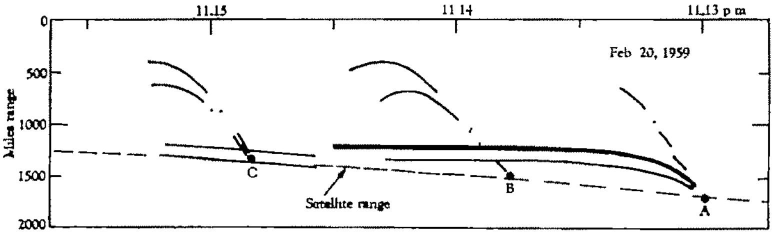

Fig. 2. Radar traces of moving ionized clouds which appear to originate at the range of an artificial Earth satellite (*Sputnik III*) as it passed through the beam of the 360-ft. parabola of the Ohio State University radio telescope

Fig. 1.) The two traces lasting about 90 sec. extending from the radiant point *A* start out with hyperbolic shape but flatten out to a constant range, suggesting that the particular clouds of particles producing them left an ionized wake nearly perpendicular to the radius vector from Columbus. From the slopes of the traces in Fig. 2, the velocities of the particle clouds radiating from *A*, *B* and *C* are between 30 and 150 km./s. These values are much in excess of the actual velocity of the satellite of about 7 km./s. Such velocities might be explained by collisions of the satellite with fast corpuscular streams. Assuming elastic collisions and an exactly head-on geometry of encounter, the resulting particle-velocity is equal to the velocity of the incident stream plus twice that of the satellite.

It should be mentioned in connexion with Fig. 2 that at about 11.13 p.m. the signals from the *Sputnik III* transmitter on 20·0045 Mc./s., which had been very strong, dropped to a very low level for about 1 min., after which their strength returned. The decrease in signal coincided with the beginning of the radar traces from the radiant point *A*, suggesting the possibility that the ionized particles encountered or produced at this time resulted in absorption or scattering of the signal from the transmitter on the satellite, while at the same time the particles acted as a reflecting or back-scattering medium for the radar signals.

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[1] Kraus, J. D., *Proc. Inst. Rad Eng.*, **46**, 611 (1958).

[2] Kraus, J. D., Higgy, R. C., and Albus, J. S., *Proc. Inst Rad Eng*, **46**, 1534 (1958).

[3] Kraus, J. D., and Dreese, E. E., *Proc. Inst. Rad. Eng*, **46**, 1580 (1958).

[4] Kraus, J. D., *Nature*, **184**, 660 (1959).

Temperatures within the Earth's Core

IN a recent communication, Strong[1] gave the results of an experimental determination of the fusion curve of iron up to a pressure of 96,000 atmospheres. On the usual assumption that the Earth's core is composed of iron, he extrapolated his results to obtain a value of 2,610 ± 200° K. for the melting point in the core at the core–mantle boundary. His experimental results fitted Simon's[2] equation:

$$\frac{p}{a} = \left(\frac{T_m}{T_0}\right)^c - 1 \tag{1}$$

with the constants $a = 75{,}000$ atmospheres and $c = 8$. In this equation p is the pressure in atmospheres, T_0 the melting temperature in degrees Kelvin at one atmosphere (that is, $T_0 = 1{,}805°$ K.), and T_m the melting temperature at the pressure p. Estimates of the melting point in the Earth's core obtained by Strong are appreciably lower than those obtained using Simon's values for the constants a and c, and only about one-half of those obtained using Gilvarry's[3] more recent values. Moreover, the gradient of the melting point curve is much lower than in these previous estimates. The value of 2,610° K. for the melting point at the core–mantle boundary is particularly interesting in view of Verhoogen's[4] conclusion that the temperature there is not likely to exceed 2,700° K. and his previous estimate[5] of 2,500° C. as the maximum, which was based on the hypothesis that the maximum observed surface temperature of lavas reflects the maximum temperature in the mantle. Since seismic evidence indicates that the outer core in any event is liquid, these two results of Strong and Verhoogen taken together fix the temperature T_1 at the core–mantle boundary rather precisely, namely:

$$2{,}610 < T_1 < 2{,}700 \tag{2}$$

so that a value of 2,650° K. for T_1 seems reasonable.

The only estimate of the melting-point temperature in the mantle is that due to Uffen[6], who obtained a value of 5,000° K. at the core–mantle boundary. With the low melting-point temperatures in the core as indicated by Strong it is worth reconsidering the early history of the Earth's interior. In my[7] explanation of the solid inner core for an Earth cooling from a molten state, it is necessary to postulate a discontinuous decrease in the melting point at the core–mantle boundary, and a higher melting point at the centre of the Earth than at the bottom of the mantle. This last requirement is certainly not met now unless Uffen's figures can be considerably reduced. A completely molten Earth would thus solidify from the core boundary and progress outwards, leaving the entire core liquid. A solid inner core could arise, however, on the hypothesis of a cold origin for the Earth, in which case melting would progress inwards from the core–mantle boundary as the temperature of the interior increased. As shown below this seems very probable.

The flatness of the gradient of the melting point curve makes it instructive to re-estimate the adiabatic gradient in the core. Vallé's estimate[8], based on seismic data and solid state theory, was greater than Simon's value of the melting point gradient and is much greater than the new values of Strong. If this is true, it would be impossible for an Earth cooling from a molten state to leave a solid inner core and a liquid outer core. Far more important, convection currents in the core which are required by most modern theories of the Earth's magnetic field, could not occur. I have thus re-computed the adiabatic gradient in the outer core, on the basis of my temperature–pressure hypothesis[9]. On this theory, the adiabatic gradient depends on the temperature and is given by:

$$\frac{\mathrm{d}T}{\mathrm{d}p} = \frac{T}{\rho\, c_p\, (C + bp)} \tag{3}$$

where ρ is the density, c_p the specific heat at constant pressure and C and b constants ($C = 2{\cdot}4 \times 10^{4}$° K., $b = 6{\cdot}2 \times 10^{-8}$° K. cm.^{2}/dyne). Equation 3 can be integrated to give:

$$\log_e T = \int \frac{\mathrm{d}p}{\rho\, c_p\, (C + bp)} \tag{4}$$

Assuming c_p to be constant[8] ($1{\cdot}24 \times 10^7$ ergs/gm. ° K.), and the temperature T_1 at the core–mantle boundary to be 2,650° K. (cf. the inequality (2)), the adiabatic temperature can be estimated at any depth within the core, and, by differentiation, the adiabatic temperature gradient. It is found that the melting-point gradient exceeds the adiabatic temperature gradient so that convection in the core would be possible. The adiabatic temperature at the boundary of the inner core estimated from equation (4) is 2,900° K. The melting point at this depth, calculated from equation (1), has exactly the same value. Although the agreement between these two estimates may well be fortuitous, it would appear that the inner core is solid.

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[1] Strong, H. M., *Nature*, **183**, 1381 (1959)

[2] Simon, F. E., *Nature*, **172**, 746 (1953).

[3] Gilvarry, J. J., *Nature*, **178**, 1249 (1956); *J. Atmos and Terr Phys*, **10**, 84 (1957).

[4] Verhoogen, J., *Trans. Amer. Geophys. Union*, **36**, 866 (1955)

[5] Verhoogen, J., *Trans Amer Geophys Union*, **35**, 85 (1954).

[6] Uffen, R. J., *Trans Amer. Geophys. Union*, **33**, 893 (1952).

[7] Jacobs, J. A., *Nature*, **172**, 297 (1953).

[8] Vallé, P. E., *Annali di Geofisica*, **5**, 41 (1952)

[9] Jacobs, J. A., *Can J Phys*, **31**, 370 (1953).

PHYSICS

Graphitization of Diamond

IN an earlier article in *Nature*[1] I reported some results of a study of the high-temperature transformation of diamond to graphite. In this work diamond powder of 0–1μ particle size was used. Samples which had been heated in vacuum to various temperatures in the range 1,000–2,000° C. were obtained and these were investigated by microscopy and diffraction in a Siemens electron microscope (Elmiskop I). Specimens which had not been heated above about 1,200° C. gave diamond diffraction patterns only, those which had been heated to 2,000° C. gave graphite patterns only, and those which had been heated to about 1,600° C. gave both diamond and graphite patterns and also some extra spots not due to either diamond or graphite. It was suggested that these extra spots were due to some intermediate arrangement of carbon atoms, possibly a regular array of diamond-like and graphite-like regions produced during the graphitization. More recent work has shown that this interpretation of the results is not justified, and it is the purpose of the present communication to describe the diffraction patterns in more detail and to give such interpretation of them as at present seems reasonable.

The transmission electron diffraction patterns were complicated, and it was not possible to obtain great accuracy in the calculated Bragg spacings because of refraction effects and other distortions of the patterns. Except for two specimens the highest spacing found was between 3·0 and 3·1 Å. The two exceptions showed reflexions at spacings of 6·0 Å. for the one and 4·1 and 3·6 Å. for the other. Lower spacings were 2·8, 2·6, 2·5, 2·4, 2·2 Å., etc., but more accurate values were afterwards obtained from X-ray powder photographs (obtained by Dr. P. Gay, Department of Mineralogy, Cambridge). These gave the values 2·60, 2·55, 2·50, 2·36, 2·18, 1·99, 1·89, 1·535, 1·419, 1·310, 1·134, 1·086 Å.

It was also found (originally by A. I. Bailey and J. W. Menter in 1950, private communication) that, if large single-crystal diamonds which had been heated to red heat in a flame were examined by reflexion electron diffraction, both regular cross-grating spot patterns and continuous ring patterns could be obtained. The spot patterns gave spacings of 8·7, 6·2, 5·8, 4·4, 3·6, and 2·5 Å. The high-spacing part of the ring patterns was lost in the shadow of the specimen edge; but rings 3·1, 2·8, 2·63, 2·52, 2·35, 2·18 Å. and lower were seen. The spacings were the same as in the transmission patterns of graphitized diamond dust. These reflexions, with the exception of those at 5·8 and 2·55 Å., can all be interpreted in terms of a cubic structure of cell side 12·3 Å. Almost all the results can thus be accounted for, and it was thought at the time that the earlier communication[1] was written that an intermediate phase must have been formed since no likely impurity having a unit cell of these dimensions exists. Furthermore, the appearance of dark-field electron micrographs suggested that the material having this structure was distributed uniformly and homogeneously through the diamond sample[2].

However, the diamond samples which gave these diffraction patterns were found by spectrographic analysis (done by Messrs. Johnson Matthey, Ltd.) to contain about 1 per cent of silicon. When the experiments were afterwards repeated using a purer diamond sample (0·0002 per cent silicon), no extra reflexions were found in the transmission electron diffraction patterns, and only one very weak extra reflexion—at 1·89 Å.—in the X-ray powder photographs. It therefore seems that a silicon compound was responsible for nearly all the extra reflexions and it is almost certain that this was silicon carbide. With the exception of the line at 1·89 Å., all the extra lines in the X-ray powder photographs agreed in spacings and relative intensities with those in a comparison pattern from silicon carbide. The spacings from most of the extra spots in the transmission electron diffraction patterns agreed with the values for silicon carbide, but there were a few exceptions—weak reflexions at 6·0, 4·1, 3·6, 3·0, and 2·8 Å. These reflexions were presumably due to some other impurity; but this has not yet been identified.

The flame-heated diamonds also gave extra reflexions when examined by reflexion electron diffraction. The ring patterns gave the same spacings as were found for the graphitized diamond dust and presumably were caused by the presence of the same impurities. The cross-grating spot patterns gave larger spacings than were found from the other patterns. They may well be due to an impurity; but again this has not yet been identified. The fact that nearly all the lattice spacings fit a cubic lattice of cell side 12·3 Å. seems to be fortuitous, especially as most of the larger spacings were only observed with the one type of specimen.

It is concluded that the hypothesis of an intermediate structure of carbon atoms is not justified, and that the extra spots and rings in the diffraction patterns were probably caused by the presence of silicon carbide and other unidentified impurities. Silicon carbide has also been found to be a contaminant of heated diamond by Raal, whose results are published in the following communication. The two sets of results are in good agreement and show the formation of silicon carbide at similar temperatures; but the source of the silicon was different—in Raal's case it was the fire-clay boat used to contain the diamond, in mine an impurity in the diamond sample

I wish to thank Dr. F. P. Bowden for his interest in this work, and Messrs. Industrial Distributors (1946), Ltd., for a grant to the Laboratory.

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[1] Seal, M., *Nature*, **182**, 1264 (1958).

[2] Seal, M., Proc. Fourth Int. Conf. on Electron Microscopy, Berlin, 1958 (in the press).

Alleged Formation of an Intermediate Diamond Structure on Heating Diamond

IT was shown by Seal[1] that little change was produced by heating natural diamond powders to 1,200° C. in a vacuum (10^{-4} mm. mercury) for periods of 15–30 min. Heating the powders to 2,000° C. resulted in a complete transformation to graphite. For temperatures of about 1,500° C. both diamond and graphite could afterwards be detected, and, in addition, the electron diffraction patterns showed extra spots not due to either diamond or graphite. Seal advanced the theory that these extra reflexions were due to some intermediate arrangement of carbon atoms in diamond, possibly a regular array of diamond and graphite-like regions.

X-ray diffraction patterns of the samples gave extra lines, by far the strongest of which corresponded with a lattice spacing of 2·51 Å. Also, it was stated that spectrographic analyses of the diamond powders before and after heating showed a chief impurity of 1 per cent silicon (Seal, M., private communication).

In a current programme of investigation diamond powders have been heated to about 1,400° C. in an inert atmosphere for various lengths of time. X-ray as well as spectrographic analyses of the powders before and after heating have confirmed the observation of Seal; but, in the light of our experiments, the existence of an intermediate atom structure in diamond after heat treatment can be discounted, and an obvious explanation for the extra X-ray reflexions and the increased silicon content of the heated diamond powder is given.

On measuring the extra lines of the X-ray pattern of the heated diamond powders the strongest reflexion was found indeed to correspond with a lattice spacing of 2·51 Å. In addition, two reasonably strong reflexions for spacings of 1·54 Å. and 1·31 Å. were found. Reference to the ASTM X-ray data card system identified these extra reflexions as being due, almost certainly, to silicon carbide.

This result is significant in the light of the fact that a chief impurity of 1 per cent of silicon is quoted by Seal for the heated diamond powders, whereas a silicon content of more than 0·1 per cent is seldom found for natural unheated diamond[3].

According to Tammann[3] the reaction between silicon and carbon to form silicon carbide starts at 1,340° C.

It appears, therefore, that the extra X-ray reflexions as well as the high silicon impurity content of the heated diamond powder are attributable to the formation of some silicon carbide as a result of chemical reaction between the diamond powder and silicon at an elevated temperature. In Seal's experiments the heating was done in crucibles of pure graphite; but the silicon might have been present initially as a contaminant or might have originated from his furnace. In our experiments, boats of fire-clay, which is essentially an aluminium silicate, were used for the heating, and it is very likely that at 1,400° C. or higher they react either with the pure diamond or a graphitized layer on the surface of the diamond to give silicon carbide.

In order to verify this a diamond powder was heated at 1,400° C. in a platinum spoon for several hours and analysed by means of X-rays as well as spectrographically. Extra X-ray reflexions were completely absent, and as can be seen from Table 1, the silicon content is not more than normally found in diamond powder.

Table 1

Diamond powder	Silicon content (percentage by weight)	Extra X-ray reflexions
Unheated	0·01	No
Heated to 1,400° C. in fire-clay boat	1·00	Yes
Heated to 1,400° C. in platinum spoon	0·04	No
Heated to 1,400° C. in fire-clay boat and treated in hydrofluoric acid	1·00	Yes
Heated to 1,400° C. in fire-clay boat and subjected to sodium-hydroxide–sodium nitrite fusion	0·01	No

A further test was done to substantiate whether, in fact, silicon carbide is formed and whether the increased silicon content of the powder heated in the fire-clay boat is not just a result of contamination from the latter.

The heated powder was treated for several hours in hot hydrofluoric acid which digests silicon in any form except as the carbide. The extra X-ray reflexions and high silicon content of the powder remained unaltered as is evident from Table 1. On subjecting this powder to a sodium hydroxide–sodium nitrite fusion which is capable of removing silicon carbide, the silicon content, as depicted in Table 1, becomes normal again and the extra X-ray reflexions are absent.

From the above considerations it is evident that the extra X-ray and electron diffraction spots observed by Seal in heat-treated diamond powders are due to the existence of easily detectable amounts of silicon carbide as a result of chemical reaction at elevated temperatures.

I wish to thank Drs. H. J. Nel and A. R. Roy for their interest and helpful discussions and Messrs. Industrial Distributors (1946), Ltd., for permission to publish this communication.

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[1] Seal, M., *Nature*, **182**, 1264 (1958).

[2] Raal, F. A., *Amer. Mineral.*, **42**, 354 (1957).

[3] Tammann, C., *Z. anorg. Chem.*, **115**, 141 (1921).

Emission and Absorption of Light behind the Detonation Front

WHEN a high explosive detonates, the measurement of the true emission of light from the detonation gases is of great interest. In air, however, the expanding detonation gas is enveloped in a compressed layer of the displaced air, which acquires a high temperature (*c.* 8,000° K. at atmospheric pressure) and emits an intense light. An easy way of eliminating the luminous layer of air is to immerse the charge in water. Charges of *TNT* and phlegmatized *PETN*, for example, then show a narrow luminous zone

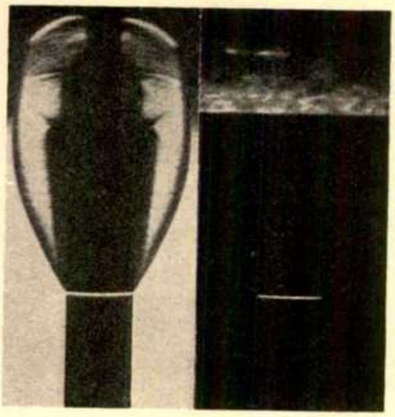
(a) (b)

Fig. 1

behind the detonation front. Fig. 1 shows instantaneous pictures (exposure time, 0·03 µsec.) of detonating cylinders of phlegmatized *PETN* of 1·55 gm./cm.^{3} density and 25-mm. diameter, first with (1*a*) then without (1*b*) background light of a detonated *TNT*-ball. (In Fig. 1*b* and Fig. 2 the upper end of the charge was above the surface.)

Fig. 1*a* shows that the expanding detonation gas is non-transparent to the background light. This implies that absorption of light is important in determining the light observed. In the case of phlegmatized *PETN* (10 per cent wax), which is very deficient in oxygen, this may be due to particles of carbon forming a light-absorbing screen on the cooled surface layer of the detonation gas. C. H. Johansson and L. Sternhoff[1] have recently shown that in *TNT*, even the narrow band of light disappears if the charge is exposed to the pressure wave from a neighbouring charge which has detonated somewhat sooner. In view of these results, the visible luminous zone cannot be assumed to correspond to the reaction zone.

In order to eliminate the absorption of light by carbon particles, similar tests were made with blasting gelatine freed from air bubbles. Instantaneous photographs show that light is emitted from a greater area with gradually decreasing intensity backwards. The diameter of the charge is, however, possibly too small for black-body emission.

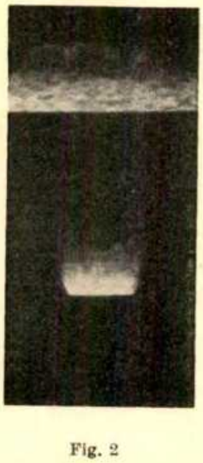
Fig. 2

To get the intense light emission of *PETN* without absorption at the surface, we have made tests with charges with a varnished core of phlegmatized *PETN* and a surface layer of blasting gelatine intended to prevent the cooling off at the surface of the detonation gases of *PETN*. The coating of varnish prevented the dissolution of the *PETN* by the blasting gelatine. Fig. 2 shows an instantaneous photograph of a detonating charge of a 25-mm. *PETN* cylinder coated with a 1-mm. layer of blasting gelatine freed from air bubbles. In this case the light is emitted over a wide area behind the front and corresponds probably much better to the temperature distribution in the expanding detonation gas of *PETN* than the narrow luminous zone in Fig. 1.

This investigation has been carried out with support of the Swedish Detonation Research Foundation.

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[1] Johansson, C. H., and Sternhoff, L., *Nature*, **183**, 247 (1959).

Effect of Corona on Travelling Waves

A NEW phenomenon of impulse corona has been observed with voltage waves of relatively long wave-fronts on a 33-kV. overhead line. The wave-shape at the sending end is 5·7/19·3 µsec. nominal. The accompanying oscillograms show typical waves recorded at a point 2·08 miles away. As the voltage exceeds the corona threshold, the wave-front shows the well-known phenomenon that part of it is sheared off due to corona losses. However, the voltage clearly recovers before its peak value is reached whereas previous observations made with wave-fronts of 1·5 µsec. or less show that this shearing effect extends to the wave tail. After the tests on which this communication is based had been completed, it was found that this recovery of voltage had also been observed by Mita and others[1] on a model line with waves of 2·2 and 4·2 µsec. fronts. As can be seen from the oscillograms, the rate of recovery is much faster for a negative wave than for a positive wave.

This phenomenon suggests that the initial intense ionization is halted, completely or almost completely, by the space charge of its own creation. This view is supported by the cessation of the high-frequency oscillation on the positive wave just before the voltage recovers. This high-frequency oscillation had also been observed to be associated with positive corona during earlier tests on 33-kV. and 132-kV. lines[2]. The fact that this recovery of voltage was not previously observed is probably due to the short front-durations of the impulse waves employed so that only the effect of the initial stage of the corona process became apparent.

Clearly, the distortion of the wave front due to corona cannot be considered to be independent of the front-duration as has been assumed in existing theory[2,3]. Either the theory has to be modified or a new approach adopted.

Several authors have measured the relation between impulse voltage $e(t)$ and corona charge $q(t)$. The results obtained by Aso[4] on a thin wire (0·6 mm. diam.) inside a concentric cylinder (10 cm. inside dia.) show that when the voltage exceeds the corona threshold dq/de increases rapidly to a fairly constant value and then decreases to a small value. It can be

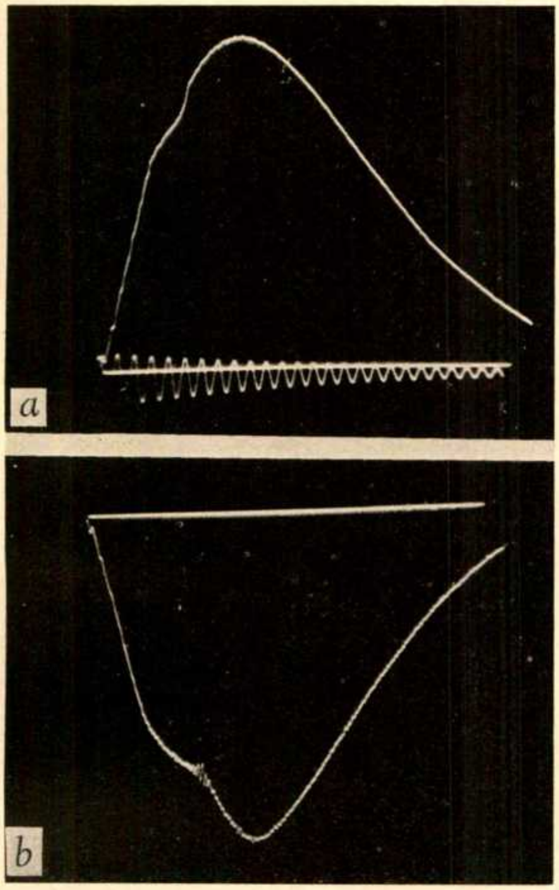

Fig. 1. *a*, Negative wave (timing oscillation, 1 Mc./s.); *b*, positive wave

shown mathematically that if the $q \sim e$ curve for the test line shows this variation in dq/de then the voltage will recover as observed.

A full report will appear elsewhere. Acknowledgment is due to the Director of the Electrical Research Association for permission to publish this communication.

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[1] Mita, N., Ikeda, G., Kinoshita, K., and Yamamori, K., *Bull. Electrotech. Lab.* (*Japan*), **21**, 125 (1957).
[2] "Surge Phenomena", The Electrical Research Association (1941).
[3] Skilling, H. H., and Dykes, P. de, *Elect. Eng.*, **56**, 850 (1937).
[4] Aso, T., *J. Inst. Elect. Eng.* (Japan), **77**, 1467 (1957).

RADIATION CHEMISTRY

Effect of Ionizing Radiation upon Xanthosine in Dilute Aqueous Solution

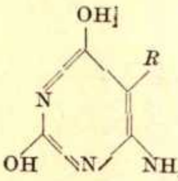
IT was reported earlier[1] that irradiation of an aqueous solution of guanosine with 15 MeV. electrons opened the imidazole ring to form 2 : 4-diamino-5-formamido-6-hydroxypyrimidine riboside. This communication reports that a similar ring-opening occurs when an aqueous solution of xanthosine is irradiated.

Table 1. R_F VALUES OF ULTRA-VIOLET ABSORBING COMPOUNDS OBTAINED FROM AN IRRADIATED AQUEOUS SOLUTION OF XANTHOSINE

Band	Solvent 1 (ref. 2)	Solvent 2 (ref. 3)	Solvent 3 (ref. 4)	Identity
1	0·46	0·31	0·67	xanthine
2	0·64	0·20	0·90	xanthosine
3	0·73	0·18	0·81	I
4	0·85	0·09	0·94	I-riboside

An aqueous solution of xanthosine, concentration 10^{-3} *M*, and containing 10^{-3} *M* neutral phosphate buffer, was irradiated with 15 MeV. electrons to a dose of 8×10^5 rads. The irradiated solution and the control solution were analysed by paper chromatography, using an 85 per cent saturated aqueous solution of ammonium bicarbonate as a solvent[2] (solvent 1). While the control solution gave a single ultra-violet absorbing band, the irradiated solution was resolved into four bands (Table 1) which were identified by their R_F's in solvents 1, 2[3] and 3[4], and the ultra-violet spectra of their eluates.

Band 1 was identified as xanthine, and is assumed to be liberated by hydrolysis following radical attack upon the ribose moiety[5]. Band 2 is unchanged solute. The spectra of bands 3 and 4 were similar with a single absorption maximum, the maximum of band 4 being at a longer wave-length (270 mμ) than that of band 3 (262 mμ). Analogy with results obtained for guanosine[1] suggested that band 3 is 2 : 6-dihydroxy-4-amino-5-formamido-pyrimidine (I) and band 4 its riboside.



I. (R = NHCHO)

II. ($R = NH_2$)

The identity of band 3 was confirmed by comparing its R_F's and ultra-violet spectra with those of an authentic specimen of I, kindly supplied by Mr. G. M. Timmis. Their R_F's in solvents 1, 2 and 3 (Table 1), and their ultra-violet spectra at *p*H 7 and 12 were identical (Fig. 1). At *p*H 1 and room temperature the ultra-violet absorption decreased slowly. In view of the well-known acid lability of formyl groups, it seemed possible that I was slowly deformylated in acid to 2,6-dihydroxy-4,5-diaminopyrimidine (II). This was essentially confirmed when, after leaving I in 0·1 *N* hydrochloric acid for 48 hr., the resulting spectra were found to be identical with those published for II[6]. Accordingly, an acid spectrum of I was obtained by dissolving it in 98 per cent formic acid (see Fig. 1), which gave no spectral change over a period of 20 hr.

Band 4 was identified as the riboside of I by identifying the sugar and ultra-violet absorbing compounds released during acid hydrolysis. Band 4 was separated from free ribose by running it in solvent 2, eluted and hydrolysed in 85 per cent formic acid at 100° C. for 5 min. When the hydrolysate was run in solvent 2 an ultra-violet absorbing product was detected with R_F's and ultra-violet spectra identical with those for I. A sugar spot, with an R_F identical with that for ribose, was detected by spraying the paper with aniline-hydrogen-phthalate[8]. Xanthosine is readily hydrolysed in acid[9]. Under the conditions

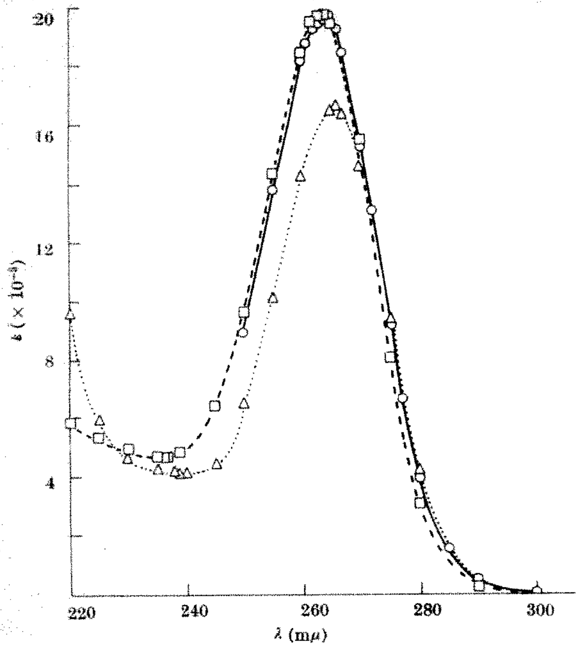


Fig. 1. Ultra-violet absorption spectra of 2 : 6-dihydroxy-4-amino-5-formamido-pyrimidine (I). ○—○, in 98 per cent formic acid ; □- - - -□, at pH 7 ; △· · · ·△ at pH 12

described above xanthosine and I-riboside (band 4) were completely hydrolysed, while under milder conditions, namely, 85 per cent formic acid at room temperature, half the starting material was hydrolysed in 22 hr. and no difference in the rates of hydrolysis of xanthosine and I-riboside was observed.

It is now established that the imidazole ring is opened when aqueous solutions of guanosine and xanthosine are irradiated. Results obtained with adenosine and inosine, to be published shortly, show that here too the imidazole ring is opened ; this seems to be a general reaction for purine nucleosides. When an electrophilic reagent approaches the purine ring C–8 appears to have the highest electron density[10]. Assuming that the chemical action of ionizing radiation is by OH radicals[11], attack at C–8 would be consistent with the electron configuration of the purine ring, mentioned above. Xanthosine is changed at a rate ($=G$) of 1·2 molecules per 100 eV. absorbed energy, which is low compared with the rate of formation of OH radicals ($G=2{\cdot}8$)[12]. Presumably the reaction is complicated by unknown factors, for example, a back-reaction, upon which the present results shed no light.

I wish to thank Prof. J. Rotblat, St. Bartholomew's Hospital Medical School, for his kind permission to use the linear accelerator. The Medical Research Council is thanked for its financial support when this work was begun at the Barnato-Joel Laboratories, Middlesex Hospital, London. Identification of products was completed at the Chester Beatty Research Institute, Institute of Cancer Research, with the financial support of the William Shepherd Fund, and by grants to the Chester Beatty Research Institute (Institute of Cancer Research: Royal Cancer Hospital) from the Medical Research Council, the British Empire Cancer Campaign, the Jane Coffin Childs Memorial Fund for Medical Research, the Anna Fuller Fund, and the National Cancer Institute of the National Institutes of Health, U.S. Public Health Service.

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[1] Hems, G., *Nature*, **181**, 1721 (1958).
[2] Hems, G., *Arch. Biochem. Biophys.*, **82**, 487 (1959).
[3] Korn, *et al.*, *J. Amer. Chem. Soc.*, **75**, 3610 (1953).
[4] Levenbrook, "The Nucleic Acids", ed. by Chargaff, E., and Davidson, J. N., **1**, footnote *j*, 252.
[5] Scholes, G., and Weiss, J., *Exp. Cell Res.*, Supp., **2**, 219 (1952).
[6] Cavalieri, L. F., Bendich, A., Tinker, J. F., and Brown, G. B., *J. Amer. Chem. Soc.*, **70**, 3875 (1948).
[7] Cavalieri, L. F., and Bendich, A., *J. Amer. Chem. Soc.*, **72**, 2587 (1950).
[8] Hanes, C. S., and Isherwood, F. A., *Biochem. J.*, **45**, 294 (1949).
[9] Levene, P. A., and Dmochowski, A., *J. Biol. Chem.*, **93**, 563 (1931).
[10] Albert, A., Ciba Symposium, 97 (1957).
[11] Weiss, J., *Nature*, **153**, 748 (1944).
[12] Allen, A. O., Hochanadel, C. J., Chormley, J. A., and Davis, T. W., *J. Phys. Chem.*, **56**, 575 (1952).

CHEMISTRY

Simple Non-gassing Electrodes for Use in Electrophoresis

THE electrodes conventionally used in electrophoresis are noble metal (usually platinum), metal/metal ion (for example, Cu/Cu^{2+}) and metal/insoluble metal salt (for example, $\text{Ag}/\text{AgCl}/\text{Cl}^-$). With the last two types the apparatus must be provided with convection barriers to prevent metal ions in the electrode compartments from contaminating the experimental material. Platinum electrodes, even if platinized, permit only a limited current density (less than 2 m.amp./cm.^{2}) without giving an evolution of gas bubbles which disturbs the observations in closed electrophoresis systems such as are used in the moving boundary and microscopic methods. We wish to describe here the general conditions under which palladium–hydrogen electrodes can be used to provide adequate current densities for electrophoresis without gassing or danger of contamination from heavy metals.

Almost a century ago Graham found that palladium metal would dissolve considerable amounts of hydrogen when used as the cathode in an electrolytic cell[1]. Present evidence indicates that the following electrolytic reactions take place at palladium electrodes[2] :

$$x\text{H}_3\text{O}^+ + \text{Pd} + xe^- = \text{PdH}_x + x\text{H}_2\text{O} \quad (\text{at low } p\text{H})$$
$$x\text{H}_2\text{O} + \text{Pd} + xe^- = \text{PdH}_x + x\text{OH}^- \quad (\text{at high } p\text{H})$$

The PdH_x formed at the cathode is not a definite chemical compound such as a hydride, but more nearly an alloy of palladium with hydrogen[3].

So long as the cathodic formation of PdH_x is fast compared with the combination reactions of atomic hydrogen on the surface of the electrode, no molecular hydrogen will be evolved until the solubility of hydrogen in palladium is exceeded at a hydrogen/palladium atomic ratio of about 0·9 (ref. 4). At the anode, oxygen or other gases will not be evolved so long as the electrode has a store of atomic hydrogen and the current density is not excessive. Obviously the anode must be cathodically charged with hydrogen before use.

While similar reactions may occur at platinized platinum electrodes, the situation is considerably altered by the fact that the total amount of hydrogen stored is limited to surface adsorption. The advantage of palladium as a current-carrying electrode lies in its capacity to absorb considerable amounts of hydrogen.

Three possible ways of using palladium electrodes are indicated schematically in Fig. 1,*a*, *b*, *c*. In the simplest system, Fig. 1,*a*, the electrodes are immersed in the solution, filling the electrode chamber. Here the anode must be precharged with an amount of hydrogen sufficient for the electrophoretic run. Relatively small electrodes can store ample amounts of hydrogen for many purposes ; for example, a palladium anode weighing 1 gm. will, if precharged to a hydrogen/palladium atomic ratio of 0·5, deliver a current of 10 m.amp. for more than 12 hr. before the hydrogen is exhausted.

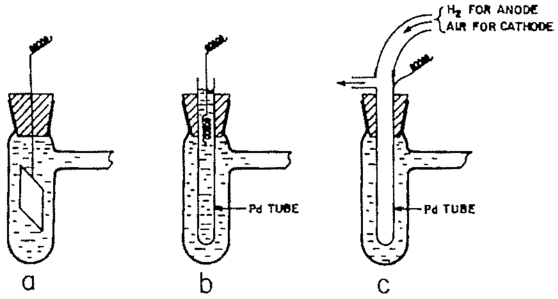

Fig. 1

We have used successfully electrodes of this type with conventional flat microelectrophoresis cells over a period of a year in studies on erythrocytes and bacteria. Current densities in excess of 10 m.amp./cm.^{2} of electrode area could ordinarily be used without gas evolution. Thus quite small electrodes are suitable even for studies at high ionic strengths.

In Fig. 1,*b*, the palladium tube serves as a bielectrode separating the solution leading to the electrophoresis cell and the solution surrounding the primary electrode (for example, Ag/AgCl). Here gas evolution resulting from exhaustion or saturation of atomic hydrogen at the electrodes would not occur regardless of the duration of the current since equivalent amounts of hydrogen are released and taken up by opposite faces of each bielectrode. The choice of the half-cell to be used inside the palladium anode tube should be made with care since the presence of soluble oxidants such as oxygen would result in the consumption of atomic hydrogen present on the palladium surface before it could enter the bulk of the metal. Hence the supply of hydrogen in the metal would eventually be depleted and gas bubbles might form at the exterior surface of the tube.

In the cell of Fig. 1,*c*, the interior of the palladium tube anode is continuously flushed with hydrogen gas and the cathode is open to the air. On passage of current through the system the hydrogen dissolved in the anode from the gas phase migrates through the wall of the palladium tube and reacts at the solution interface as indicated in the equations given above. At the cathode, atomic hydrogen derived from the reduction of hydronium ions in the solution migrates through the wall of the tube and reacts with atmospheric oxygen to form water. With these electrodes, current could obviously be passed in one direction indefinitely with no danger of gas formation at either electrode. This system would not be convenient, however, where frequent reversal in current direction is necessary (as in microelectrophoresis) since the hydrogen and air connexions would have to be exchanged with each reversal. Electrodes of this type have been used successfully in an electrochemical cell[6].

A few precautions in the use of palladium electrodes are worth mentioning. A palladium anode immersed in a halide solution will react to give a soluble palladium halide salt if its store of hydrogen is exhausted. Charging a palladium electrode beyond a hydrogen/palladium ratio of about 0·6 will lead to an increase in volume of about 10 per cent[7]. Repeated cycles of charging beyond this ratio followed by complete discharge may cause the metal to crack. By restricting the maximum amount of hydrogen dissolved to a ratio of about 0·5 the electrodes may be expected to have an indefinite life.

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[1] Graham, T., *Proc. Roy. Soc.*, 15, 223 (1866); *ibid.*, 16, 422 (1868).
[2] Hoare, J. P., and Schuldiner, S., *J. Electrochem. Soc.*, 102, 485 (1955)
[3] Mott, N. F., and Jones, H., "The Theory of the Properties of Metals and Alloys", 190–200 (Clarendon Press, Oxford, 1936).
[4] Kruger, F., and Gehm, G, *Ann. Physik*, (5), 16, 174 (1933).
[5] Hoare, J. P., and Schuldiner, S., *J Electrochem. Soc*, 104, 564 (1957).
[6] Schuldiner, S., *J. Electrochem. Soc.*, 106, 440 (1959).
[7] Smith, D. P., "Hydrogen in Metals" (Univ Chicago Press, Chicago, 1948).

Separation of Uranium by Reversed Phase-Partition Chromatography

IN almost all analytical methods for the determination of uranium, it is necessary first to separate the uranium from other substances.

A convenient and selective method of achieving this separation is to extract the uranium from a nitric acid solution into a solution of tributyl phosphate in kerosene, the latter serving to facilitate separation of the two phases. Usually two extractions are required, one to transfer uranium to the organic phase and one to return it to an aqueous phase for determination.

The use of tributyl phosphate on a partition chromatographic column would reduce the hazard of manipulating radioactive material, improve the partition of uranium into the organic phase, and have the effect of a large number of manual solvent extractions. Such a column has been successfully used in these laboratories for the past eleven months (internal publication and patent application).

The stationary phase is prepared from purified tributyl phosphate. A mixture of 100 ml. of commercial tributyl phosphate and 150 ml. of 0·5 *M* sodium hydroxide is steam distilled until no odour of butyl alcohol is present in the distillate. After cooling, the residual tributyl phosphate is separated from the alkali and washed with 100 ml. of distilled water, followed by three 50-ml. portions of 3 *M* nitric acid. For the solid support, 'Low density, *Kel-F* 300 moulding powder' (Minnesota Mining and Manufacturing Co.) is ground, using solid carbon dioxide to cool the material, and the powder passing 100 B.S.S. and retained by 200 B.S.S. is collected. The column is prepared by adding 3 ml. of purified tributyl phosphate to 3 gm. of the '*Kel-F*' powder, mixing with a glass rod, and loading as a slurry in 5·5 *M* nitric acid into a burette 7 mm. in diameter containing a loose plug of cotton-wool above the tap. The slurry is agitated with a glass rod to release air bubbles, and 5·5 *M* nitric acid is run through the column to consolidate the contents. A loose plug of cotton-wool is placed over the material to prevent disturbance of the surface.

The sample solution, 5–10 ml., containing not more than 50 mgm. of uranium in 5·5 *M* nitric acid, is poured on to the column. The uranium is retained as a clearly defined yellow band at the top of the column and impurities are eluted by washing with three 10-ml. portions of 5·5 *M* nitric acid at a rate of flow of 1–2 ml. per min. The uranium is then eluted with 35 ml. of distilled water at the same rate and determined by a suitable method.

The column is reactivated by washing with 20 ml. of 5·5 *M* nitric acid. The tributyl phosphate is slowly eluted, but one column packing can be used for more than fifty determinations, and the *Kel-F* from exhausted columns may be used as support material after washing with carbon tetrachloride, and drying.

Most impurities are eluted rapidly, the peak of the elution being reached when 10–12 ml. of eluent have passed through the column. The peak for the elution of uranium with nitric acid is not reached until 200 ml. of eluent have passed through and there is a clear volume of about 40 ml. between the two peaks. In order to save time, the eluent is changed to water after the impurities have been eluted.

Uranium has been separated effectively from more than ten times its weight of chromium, copper, aluminium, nickel, vanadium, calcium, iron, molybdenum, tungsten, sodium, potassium, fluoride, phosphate, sulphate, chloride and from concentrated solutions of citric acid which complex uranium strongly. The method is applicable from the microgram- to the gram-level by proportional adjustment of the size of column. Recoveries at the level of 5 mgm. of uranium have been in excess of 99 per cent, which has been adequate for the purpose in hand. We are confident, however, that a quantitative recovery is possible, and the method is being further investigated. A full report of the work will be published.

The tributyl phosphate/nitric acid column has been used to separate uranium from some of its daughter products.

Thorium, plutonium IV and cerium IV are retained on the column after washing with nitric acid, but cerium can be separated after reduction by sulphur dioxide to cerium III.

By modifying the technique so as to use hydrochloric acid in place of nitric acid throughout, it has proved possible to separate plutonium from uranium The plutonium must be reduced to the trivalent form before loading on to the column. The uranium is retained on the column.

We are grateful to the managing director of the Production Group of the United Kingdom Atomic Energy Authority for permission to publish this communication.

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Evidence for the Existence of a Potassium Rhenium Hydride and its Bearing on the Nature of the (−1)-Oxidation State of Rhenium

THIS is a preliminary report of some observations we have made on the product of the reduction of potassium perrhenate with potassium metal in ethylenediamine–water solution. According to Bravo, Griswold and Kleinberg[1], and also Floss and Grosse[2], rhenium is reduced to the (−1)-oxidation state in this reaction, and the solid rhenide $KRe.4H_2O$ may be isolated from the products.

The reduction procedure and method of separating the unreacted potassium perrhenate ($KReO_4$) and the bulk of the potassium hydroxide from the product were as described by Bravo, Griswold and Kleinberg[1]. The grey-brown solid obtained in this manner was purified by dissolving it in an alkaline 10 : 2 methanol–water solution, centrifuging to settle out the brown-black solid matter (rhenium metal or a rhenium oxide), and then slowly adding the clear supernatant to a large excess of oxygen-free, anhydrous *iso*propanol. The result is a white precipitate which was successively extracted three times with *iso*propanol and then three times with anhydrous ethyl ether; after which it was pumped down for 2 hr. at 90° C. This product is stable in an inert atmosphere.

On treating the white solid or its solution in aqueous alkali (which is quite stable) with various oxidizing agents (hypochlorite, thallous ion, permanganate) gas evolution takes place. The gas evolved when a solution of the solid in 5 *M* potassium hydroxide reacted with 0·1 *M* potassium permanganate was shown, by mass-spectrometric analysis, to be pure hydrogen. The volume of hydrogen evolved on treating about 50-mgm. samples of the solid with 25 ml. of a solution 4·5 *N* in alkali and 1 *N* in permanganate is about 10 ml. at room temperature and atmospheric pressure. We find, as the mean of three measurements:

gram atoms H evolved/gram atoms Re $= 5 \cdot 54 \pm 0 \cdot 06$

In acid solution the solid decomposes with the evolution of hydrogen and the precipitation of rhenium metal. Treatment of about 55-mgm. samples with excess of dilute sulphuric acid yields about 20 ml. of hydrogen at room temperature and atmospheric pressure. As the mean of three measurements we find:

gram atoms H evolved/gram atoms Re $= 10 \cdot 05 \pm 0 \cdot 05$

The above results indicate that the white solid contains a hydride of rhenium. To confirm this, about 40 mgm. of the solid were dissolved in about 0·2 ml. of de-aerated 50 per cent potassium hydroxide solution, and the proton resonance spectrum was obtained with a high-resolution nuclear magnetic resonance spectrometer operating at a frequency of 60 Mc./s. The spectrum consists of two peaks separated by 14·7 parts per million. The low-field peak is very large and is due to the water–hydroxyl ion protons. The high-field peak is much smaller and can only be attributed to protons of a hydridic nature (H^-). The shift of the hydride peak with respect to the proton resonance of pure water is + 13·3 p.p.m. Wilkinson and co-workers have independently obtained a similar spectrum for the solution resulting from the reaction of aqueous potassium perrhenate with sodium amalgam (Wilkinson, G., personal communication). In addition, we have observed the proton resonance spectrum of the solid dissolved in deuterium oxide (heavy water) containing about 35 per cent sodium deuteroxide (NaOD) (no proton resonance could be observed on the solvent). Again the spectrum consists of only two peaks separated by 14·7 p.p.m. This shows that the solid contains H_2O or OH^- groups as well as hydridic protons. The fact that only two peaks occur indicates that alcohol and ethylenediamine are absent, and this is confirmed by chemical tests. The ratio of the intensity of H^- peak/intensity of $H_2O - OH^-$ peak $\simeq 2$. Hence the ratio H^-/H^+ in H_2O or $OH^- \simeq 2$; but it must be noted that the sample used in the deuterium oxide experiment had been standing under vacuum for about one month and may have been partially dehydrated.

These observations demonstrate the existence of a soluble rhenium hydride. This is the first example of a soluble transition metal hydride which does not also contain complexing groups such as $C_5H_5^-$, CO or CN^- attached to the metal ion (bis*cyclo*pentadienylrhenium hydride and rhenium carbonyl hydrides are known[3]). We believe that it is now no longer necessary to postulate the existence of the rhenide ion, Re^-, or an aquo complex of this ion, since all the properties of the apparent (−1)-oxidation state of rhenium[1,2,4] may be explained as being due to a hydride of a positive oxidation state of rhenium. Work on this problem is continuing, and it appears possible that more than one rhenium hydride may exist. Further details will be reported at a later date.

This work was supported in part by the U.S. Atomic Energy Commission.

Note added in proof. Two measurements of the area beneath the high-field proton-resonance peak in deuterated basic aqueous solvent gave 7·3 and 6·5 for the ratio of hydridic hydrogen atoms to rhenium atoms in the compound. The measurement, expected to be reliable to about 20 per cent, was calibrated by the addition of a known quantity of normal-isotopic water to the deuterated solvent.

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[1] Bravo, J., Griswold, E., and Kleinberg, J., *J. Phys. Chem.*, **58**, 18 (1954).

[2] Floss, J. G., and Grosse, A. v., *J. Inorg. Nuclear Chem.*, **9**, 318 (1959).

[3] See, for example, Wilkinson, G., and Birmingham, J. M., *J. Amer. Chem. Soc.*, **77**, 3421 (1955).

[4] Lundell, G. E. F., and Knowles, H. B., *J. Res. Nat. Bur. Stand.*, **18**, 629 (1937); Tomicek, O., and Tomicek, F., *Coll. Czech. Chem. Comm.*, **11**, 626 (1939). Lingane, J. J., *J. Amer. Chem. Soc.*, **64**, 1001, 2182 (1942). Maun, E. K., and Davidson, N., *ibid.*, **72**, 3509 (1950). Rulfs, C. L., and Elving, P. J., *ibid.*, **73**, 3284, 3287 (1951).

Formation of Volatile Compounds on Gas–Liquid Chromatography Columns

THE communication by Weurman and Dhont[1] reporting the formation of volatile esters on gas–liquid chromatography columns using polyethylene glycol 600 prompts us to report our own findings with a similar stationary liquid phase.

When first preparing a column filling with polyethylene glycol 400, it was distributed on 'Celite 545' in a mixed solvent of acetone and diethyl ether which was afterwards removed by heating in an oven at 100° C. for 3 hr. During this period a strong smell of formaldehyde was noticed but this later disappeared, and the column was therefore packed and brought into use at 60° C. with nitrogen as carrier gas. The copper connexions of the thermistor detector became corroded and the base line irregular. The stock of polyethylene glycol 400 was afterwards tested and found to contain formaldehyde (chromotropic acid test) and formic acid (paper chromatogram by Dr. G. C. Whiting).

Since this time we have pre-treated the polyethylene glycol 400 as suggested by Adlard[2], namely, by heating to 100° C., but under a higher vacuum (0·01 mm. of mercury), for 16 hr. Satisfactory results have since been obtained, chromatograms of alcohols showing no abnormalities. It thus seems that polyethylene glycols as supplied by the manufacturers contain volatile impurities which should be removed before use as stationary phases. Our findings offer an explanation of the source of formic acid causing the esterification observed by Weurman and Dhont.

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[1] Weurman, C., and Dhont, J., *Nature*, **184**, 1480 (1959).

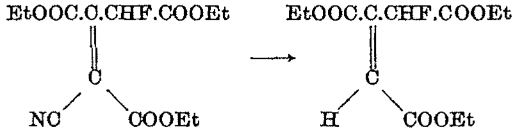
[2] Adlard, E. R., "An Evaluation of Some Polyglycols used as Stationary Phases for Gas-Liquid Partition Chromatography" Symposium on Vapour Phase Chromatography, Institute of Petroleum, London, May–June, 1956.

Triethyl α-Fluoroaconitate

THE current theory on the mode of action of fluoroacetate[1] implies the lethal synthesis of fluorocitrate which poisons the enzyme aconitase, thus preventing the catabolism of citrate to aconitate; it seemed of interest to prepare and study α-fluoroaconitic acid, HOOC.CHF.C(COOH)=CH.COOH. Triethyl fluorocitrate, which would appear to be a suitable starting material, has been prepared in a facile reaction, namely, by the Reformatzky reaction between diethyl oxalofluoroacetate and ethyl bromoacetate[2], but our attempts to dehydrate this compound failed to give a defined product.

Better results were obtained by the condensation of diethyl oxalofluoroacetate with ethyl cyanoacetate in the presence of piperidinium acetate. The product so obtained (I) gave, upon treatment with alcoholic sulphuric acid, directly the desired triethyl α-fluoroaconitate (II).

$\mathrm{EtOOC.CO.CHF.COOEt} + \mathrm{NC.CH_2.COOEt} \rightarrow$



$$\begin{array}{c}\mathrm{EtOOC.C.CHF.COOEt}\\ \| \\ \mathrm{C} \\ \mathrm{NC}\diagup\quad\diagdown\mathrm{COOEt}\end{array} \longrightarrow \begin{array}{c}\mathrm{EtOOC.C.CHF.COOEt}\\ \| \\ \mathrm{C} \\ \mathrm{H}\diagup\quad\diagdown\mathrm{COOEt}\end{array}$$

I II

We have not succeeded in hydrolysing (II) without far-reaching degradation of the molecule. However, the biological properties of (II) as such will be of interest, as the complications inherent in the possible stereochemical forms of fluorocitrate[3] are not to be feared in (II).

Triethyl γ-cyano-α-fluoroaconitate (*I*). A mixture of 21 gm. of diethyl oxalofluoroacetate[4], 14 gm. of ethyl cyanoacetate, 0·5 ml. of acetic acid and 0·1 ml. of piperidine was heated for 20 hr. at 100° in a vacuum of 35 mm. After 6 and again after 13 hr. of this period, 0·05 ml. of piperidine and 0·25 ml. of acetic acid were added. The product was subjected directly to distillation under 0·05 mm. pressure. The ester (I) boiled at 160–162° and formed a colourless oil; yield, 12 gm. (40 per cent).

Analysis: Calculated for $C_{13}H_{16}NO_6F$: C, 51·8; H, 5·3; N, 4·7; F, 6·3.

Found: C, 51·5; H, 5·6; N, 4·3; F, 5·9.

Triethyl α-fluoroaconitate (*II*). A mixture of 5 gm. of concentrated sulphuric acid and 1 gm. of water was added to a solution of 12 gm. of (I) in 20 ml. of anhydrous alcohol, refluxed for 5 hr. and poured into cold 10 per cent sodium hydrogen carbonate solution. Extraction with ether, drying and distillation gave 5 gm. (45 per cent) of triethyl α-fluoroaconitate (II), b.p. 115–117° (0 1 mm.).

Analysis: Calculated for $C_{12}H_{17}FO_6$: C, 52·2; H, 6·2; F, 6·9.

Found: C, 52·4; H, 6·3; F, 6·5.

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[1] Peters, R. A., "Adv. in Enzymol.", 18, 13 (Interscience Pub. Inc New York, 1957)

[2] Rivett, D. E. A., *J. Chem. Soc.*, 3710 (1953).

[3] Kaeser, H., *Discuss. Farad. Soc.*, **20**, 289 (1955)

[4] Blank, L., Mager, J., and Bergmann, E. D., *Bull. Res. Council Israel*, **3**, 101 (1953), *J. Chem. Soc.*, 2180 (1955)

Colorimetric Determination of Amino-acid Concentration

A VERSATILE method of amino-acid determination is that in which amino-acid solutions are incubated with cupric orthophosphate in suspension. The resulting solution contains copper in proportion to the total molarity of amino-acids and the copper is determined by any suitable method. The methods used have included iodometric titration[1] and the use of cupric phosphate labelled with copper-64 followed by the determination of the activity in the supernatant[2]. The latter method is of particular use for the continuous determination of amino-acids in chromatographic eluates.

I have determined the copper colorimetrically using sodium diethyl dithio carbamate as the copper reagent. The colour produced is stable and has a maximum absorption at 440μ. The stages in the procedure are as follows.

(1) 0·1 ml. of the sample, 3·0 ml. of cupric orthophosphate suspension (1 per cent aq.), 2·0 ml. of B.D.H. Universal buffer solution, *p*H 7·0[3] are incubated overnight in a centrifuge tube. A reagent blank is used with distilled water replacing the sample. Using finely divided cupric orthophosphate (B.D.H.) shaking was found to be unnecessary even when using conical centrifuge tubes, the recovery figures being identical in procedures with and without shaking. The tubes are centrifuged to sediment the cupric orthophosphate and a part of the supernatant withdrawn for colour development.

(2) 0·1 ml. of the supernatant from (1), 3 0 ml. of B.D.H. Universal buffer *p*H 7·0, 0·1 ml. of sodium diethyl dithio carbamate 1·0 per cent aq., are mixed at room temperature. The optical density at 440μ does not increase after 15 min. from mixing, and is read at any convenient time, being stable for at least 24 hr. A 'Unicam' *S.P.* 500 or similar instrument is used with a violet filter. If the colour is too dense or if precipitation occurs, the supernatant from stage (1) is diluted and stage (2) repeated. The results obtained show that, due to the large excess of cupric orthophosphate in stage (1), the relationship [copper]/[amino-acid] is linear over a wide range, so that dilution is permissible with no inherent inaccuracy. The 'recovery' of added amino-acids was 100 per cent ± 2 per cent over a concentration range of 50–1,000 μgm./ml. α-alanine. This method has proved to be useful in enzyme kinetic work.

Nielsen[4] describes the estimation of proteins by a related method, involving the removal of amino-acids before reaction of the proteins and trichloracetic acid-insoluble peptides with cupric sulphate. Excess copper is removed by precipitation as cupric orthophosphate and the copper in solution as a peptide complex is estimated using diethyl dithio carbamic acid. In the method here described, peptides and proteins are found not to give any appreciable copper uptake from cupric orthophosphate, the results for amino-acid concentration obtained from solutions containing amino-acids, peptides and proteins being within 2 per cent of those obtained by formol titration and by the Folin–Ciocalteau method[5].

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[1] Pope, C. G., and Stevens, M. F., *Biochem. J.* **33**, 1070 (1939)

[2] Blackburn, S., and Robson, A., *Biochem. J.*, **54**, 295 (1953).

[3] Prideaux, E. B. R., and Ward, A. T., *J. Chem. Soc.*, **125**, 426 (1924).

[4] Nielsen, H., *Acta Chem. Scand.*, **12**, 1, 38 (1958).

[5] Wallenfels, K., *Biochem. Z.*, **321**, 189 (1950)

BIOCHEMISTRY

Volatile Amines in Brain

WE have reported previously the isolation and identification of the volatile amine 2-dimethylaminoethanol in the brain of man and pig and in other biological material[1,2]. Bremer and Greenberg's[3] report on its presence in rat-liver phospholipids has substantiated our findings. During our investigation several volatile amines—besides the previously identified volatile bases ammonia[4] and 2-aminoethanol[5]—were isolated from brain tissue and we became interested in analysing the brain for other normally occurring volatile amines.

Brain tissues from cat, pig and man, processed 1, 30 and 360 min. after death, respectively, were used. The extraction was carried out by three different methods to get a more accurate picture of the amines present. Accordingly, the tissue was extracted 3–4 times with either 80 per cent ethanol, adjusted to *p*H 2 with hydrochloric acid (EtOH–HCl), as described earlier[2]; or with acetone followed by 80 per cent ethanol (acetone–EtOH); or with 5 per cent trichloracetic acid. The supernatant contained the unbound or free amines; the precipitate, the bound amines. Since we were primarily interested in the unbound amine content of the brain, most of the following procedures involved only the supernatant fractions. In the case of the trichloracetic acid treatment the aqueous solution was extracted with ether and passed through an anion exchange column ('Dowex 2', Cl⁻ form), in order to remove the trichloracetic acid. The supernatants were individually concentrated *in* *vacuo* and subjected to a fractional steam distillation in saturated barium hydroxide solution or, in the case of the trichloracetic acid extract, in saturated sodium carbonate solution. The distillation and the subsequent procedures, used for obtaining the volatile amine fractions as hydrochlorides, were the same as previously reported[3]. To liberate the bound amines from human and pig brain tissue the (EtOH–HCl) precipitate was hydrolysed in saturated aqueous barium hydroxide and the bases fractionated according to the procedure described[3].

The different fractions were analysed by gas-chromatography, paper-electrophoresis and paper-chromatography. Gas-chromatography was performed with argon as the carrier gas and an ionization detector which has a usable sensitivity of 10^{-10}–10^{-11} mole. The insensitivity of the ionization detector to water allowed us to analyse the free amines in aqueous solution. The free amines were released from their hydrochloride salts with 2 *N* aqueous sodium hydroxide and injected into the gas-chromatograph, where they were separated by a carbowax-on-alkaline-chromosorb column[4]. The paper-electrophoretic and paper-chromatographic separations were carried out as previously described[5], using the hydrochloride salts of the amines.

The gas-chromatograms of the unbound amine fractions from the EtOH–HCl and acetone–EtOH extracts of pig brain and the trichloracetic acid extract of cat brain, as well as the bound amine fractions from pig brain (EtOH–HCl), showed quite similar patterns. 2-Dimethylaminoethanol, 2-aminoethanol and ammonia were present in these fractions plus 4–8 as yet unidentified amines. One of these amine-peaks recurs in the gas-chromatograms of all specimens and indicates the presence of 2-methylaminoethanol. Extracts from a human brain (EtOH–HCl) showed in the unbound and bound amine fractions an additional peak in large amounts which could be identified as 2-diethylaminoethanol.

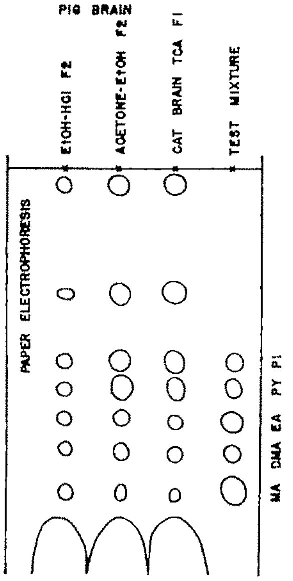

Fig. 1. Paper electrophoretic separation of brain fractions and test mixture. The migration values (where cysteine has a migration value of 0 and methylamine of 1) and dye reactions of the amines are methylamine (MA) 1·0, ninhydrin red-violet; dimethylamine (DMA) 0·90, ninhydrin red-violet; ethylamine (EA) 0·79, ninhydrin blue-violet; pyrrolidine (PY) 0·69, ninhydrin yellow, isatin blue; piperidine (PI) 0·50, ninhydrin violet, red fluorescence under ultra-violet light

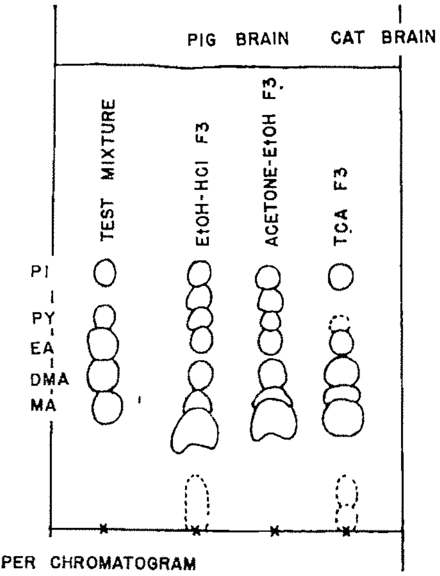

Fig. 2. Paper chromatographic separation of brain fractions and test mixture. The migration values are: methylamine (MA) 0·27; dimethylamine (DMA) 0·32; ethylamine (EA) 0·40; pyrrolidine (PY) 0·44; piperidine (PI) 0·53. The dye reactions were the same as in Fig. 1

It was learned later that the patient whose brain was analysed received treatment with 110 mgm. of procaine (in the form of procaine penicillin) every 6 hr. prior to death. This probably accounts for the large amount of 2-diethylaminoethanol present. Also, the occurrence of this substance in the bound form suggests the possibility of its incorporation into phospholipids.

Figs. 1 and 2 represent the paper-electrophoretic and paper-chromatographic separation of the pig and cat brain fractions, obtained by the different extraction procedures. It can be seen that these fractions are almost identical. By comparison with known amines, five of the spots could be identified as methylamine, dimethylamine, ethylamine, pyrrolidine and piperidine. These five substances were present in the free amine fractions of the EtOH–HCl and acetone–HCl extracts from pig brain and in the free amine fractions from cat brain, obtained by trichloracetic acid extraction. In the bound fractions from pig brain (EtOH–HCl) methylamine, dimethylamine, ethylamine and pyrrolidine were identified. The human brain (EtOH–HCl extraction) contained in the free amine fractions methylamine, dimethylamine, ethylamine and piperidine; in the bound amine fractions piperidine was missing. The migration values and dye reactions of the substances from the brain extracts and the mixture of reference compounds coincided in the chromatographic and electrophoretic systems. In addition, the reference compounds were mixed with the brain fractions and the chromatographic and electrophoretic concordance was determined from the increased spot intensities. The results were further substantiated by a two-dimensional electrophoretic-chromatographic separation on one paper[6]. Besides the identified amines this separation revealed a substantial number of unidentified amines. The largest number was found in the EtOH–HCl extract of pig brain, 12 unknown spots in the free amine and 11 in the bound amine fractions. This might be due to the relatively large amount of tissue analysed, which allowed detection of amines present in low concentrations.

Table 1. APPROXIMATE AMOUNTS OF UNBOUND VOLATILE AMINES IN BRAIN (IN FREE AMINE FORM)

	Pig brain		Cat brain
	EtOH–HCl (μgm./kgm.)	Acetone–EtOH (μgm /kgm)	TCA (μgm./kgm.)
Methylamine	110	40	790
Dimethylamine	550	290	2,460
Ethylamine	90	60	360
Pyrrolidine	20	10	20
Piperidine	30	180	840
Dimethylamino-ethanol	45	40	80

Table 1 gives the approximate amounts of the volatile amines which we found in brain tissue. The value for 2-dimethylaminoethanol is most exact because it was calculated from the peak which appeared on the gas-chromatograms, whereas the remaining substances have been estimated from the density of the spots as they appeared in paper-chromatography and paper-electrophoresis. Furthermore, since much more pig brain than cat brain was assayed, the figures for pig brain may be more representative than those for the cat brain.

Further work is planned to confirm these results with methods which exclude possible hydrolysis of unstable substances and to learn more about the significance of the occurrence of these substances in the various organs of animals under varying physiological conditions.

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[1] Honegger, C. G., *Fed. Proc.*, 18, 408 (1959).

[2] Honegger, C. G., and Honegger, Ruth, *Nature*, 184, 550 (1959).

[3] Bremer, J., and Greenberg, D. M., *Biochim. Biophys. Acta*, 35, 287 (1959).

[4] Schwarz, H., and Dibold, H., *Klin. Wschr.*, 10, 553 (1931). Riebeling, C., *ibid.*, 10, 554 (1931).

[5] Mueller, E., *Z. Biol.*, 100, 249 (1940).

[6] Honegger, C. G., *Helv. Chim. Acta*, 40, 846 (1957).

Methanesulphonic Acid Derivatives of Central Stimulant Drugs

AMONG the substances recently introduced in the therapy of mental depression which stimulate the central nervous system, iproniazide (1-*iso*nicotinoyl-2-*iso*propyl-hydrazine) has been the drug longest under investigation[1]. Its stimulating properties have been discovered by studying this drug as a tuberculostatic agent; and during this work toxic psychoses have been observed as side-effects.

A great number of hydrazine derivatives tested afterwards reacted in the same way and showed a more or less pronounced inhibition on the monoamine oxidase, an enzyme involved in the metabolism of serotonin, epinephrine and other amines. These substances unfortunately produce a wide variety of side-effects, so that many physicians refuse to use them.

Table 1. TOXICITY (*LD*50)

Drug	Administered orally (rats) (mgm./kgm.)	Administered intravenously (mice) (mgm./kgm.)
(1) Iproniazide	365	820
(2) 2-Hydrazino-propane hydrochloride	80	315
(3) 1-Phenyl-2-propyl-hydrazine hydrochloride	13	—
(4) Sodium salt of methane-sulphonic acid of (2)	330	500
(5) Sodium salt of methane-sulphonic acid of (3)	56	—

Table 2. INHIBITION *in vivo* OF THE MONOAMINE OXIDASE
Dose of drug producing 50 per cent inhibition, administered endoperitoneally 6 hr. before the animals were killed (ref. 6)

Drug	Brain (mgm.)	Liver (mgm.)
(1) Iproniazide	6 2	1 2
(2) 1-Phenyl-2-propyl-hydrazine hydrochloride	0·4	0 45
(3) Sodium salt of methane-sulphonic acid of (2)	0·70	0·70
(4) Sodium salt of methane-sulphonic acid of 2-hydrazino-propane	2·45	0·61

Table 3. BRAIN SEROTONIN-LEVELS OF RATS AFTER ADMINISTRATION OF DRUGS 6 HR. BEFORE THE ANIMALS WERE KILLED (REF. 6)

Drug	Dose (mgm./kgm.)	No. of animals	Brain serotonin (γ/gm. fresh weight ± S.E.)
Control	—	15	0·470 ± 0 021
(1) Iproniazide	100	10	0·994 ± 0 038
(2) Sodium salt of methanesulphonic acid of 2-hydrazino-propane	114	10	1·400 ± 0·045
(3) 2-Hydrazino-propane hydrochloride	61	10	1·860 ± 0 065

In order to get a therapeutically more suitable drug with a lower toxicity we adopted a method which has already given good results with other drugs[6], namely, transformation into methanesulphonic acid derivatives. Thus we transformed 2-hydrazino-propane[3] and 1-phenyl-2-hydrazino-propane[4] (both substances are considerably more active than iproniazide) into the above derivatives. We treated 2-hydrazino-propane and 1-phenyl-2-hydrazino-propane with formaldehyde (40 per cent) and then with sulphur dioxide to obtain the corresponding methanesulphonic acids, which we neutralized with sodium bicarbonate to form the sodium salt of methanesulphonic acid of 2-hydrazinopropan (I) and the sodium salt of methanesulphonic acid of 1-phenyl-2-hydrazino-propane (II).

The sodium salts have no definite melting point, but the acid corresponding to (I) melts at 158°, that of (II) at 136–138° [5]. Analysis of the acid of (I) gave:

Calculated for $C_4H_{12}N_2SO_3$: C, 28·55; H, 7·19; N, 16·42; S, 18·50.

Found: C, 28·33; H, 7·16; N, 16·42; S, 18·50.

Analysis of the sodium salt of (I) gave:

Calculated for $C_4H_{11}N_2SO_3Na.2H_2O$: N, 12·37.

Found: N, 12·85.

Analysis of the sodium salt of (II) gave:

Calculated for $C_{10}H_{15}N_2SO_3Na.3H_2O$: N, 8·76; S, 10·02.

Found: N, 8·40; S, 9·82.

Table 1 shows a lower toxicity compared to the initial substances, and Tables 2 and 3 show in the same time a high activity with regard to the inhibition of monoamine oxidase and to the increase of the brain serotonin.

Th. Bersin[7] has attributed this surprising change of the pharmacological properties of the methanesulphonic acid derivatives to the transformation of a cationic drug into an anionic one:

$$R\text{NHNH}_3^+ \rightarrow R\text{NH.NH.CH}_2\text{SO}_3^-$$

This transformation presumably changes the penetration and excretion of the drug.

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[1] See *J. Amer. Med. Assoc.*, 168, 286 (1958).

[2] See conclusion, Logemann, W., and Miori, G. P., *Arzneimittelforschung*, 5, 218 (1955).

[3] Pletscher, A., and Gey, K. F., *Helv. Physiol. Pharmacol. Acta*, 16, 026 (1958).

[4] Biel, J. H., Drukker, A. E., Shore, P. A., Spector, S., and Brodie, B. B., *J. Amer. Chem. Soc.*, 80, 1519 (1958).

[5] This acid has recently been obtained by another method; see Biel, J. H., Drukker, A. E., Mitschell, Th. F., Sprengeler, E. P., Nuhfer, P. A., Conway, A. C., and Horita, H., *J. Amer. Chem. Soc.*, 81, 2805 (1959).

[6] For further details see Manzini, B., Goldwurm, G. F., and Canal, N., *Boll. Soc. Ital. Biol. speriment.*, 34, 1407 (1958). Canal, N., Maffei-Faccioli, A., and Manzini, B., *ibid.*, 34, 1525 (1958); and *Atti Soc. Lomb. Scienze Med. e Biol.* (in the press).

[7] Bersin, Th., in "Ion Exchangers in Organic and Biochemistry", 498 (Intersci. Publ., New York, 1957).

Release of Iron from Conjugates in Foods

IRON in food of both animal and plant origin must be released from its conjugates before it can be absorbed from the gastro-intestinal tract. There is also evidence that it must be in the reduced ferrous state before assimilation. The total iron and the percentage easily available as ferrous iron are therefore both important in assessing the dietary value of an iron-containing food, and for this reason most tables of food values show the total iron and that amount which reacts with α-α′-dipyridyl. These measurements, however, do not recognize the possible effects of cooking and of intestinal enzymatic digestion in releasing iron from its conjugates.

Kaldor demonstrated that 12–13 per cent of the total iron in hæmoglobin is liberated by acid peptic digestion of heat-denatured blood, whereas only approximately 0·1 per cent could be extracted with saline from uncooked blood[1]. Brading, Kaldor and George also showed that more iron was absorbed by rats from cooked than from uncooked blood administered into the stomach[2]. These observations have been based solely on hæmoglobin, although Moore and Dubach have shown that iron-59 incorporated into various foods is absorbed from the gastro-intestinal tract[3]. The part played by acid peptic digestion *in vitro* in liberating iron from cooked foods has now been extended to a number of other materials.

The total iron content was determined on acid digests of the foods. Aliquots of calves' liver and flour were heated to 95° C. for 30 min., and of silver beet and sultanas to 100° C. for 30 min. They were then homogenized in the water or saline in which they had been heated. One aliquot of each homogenate was incubated with water or saline at 37° C. for 60 min. and another with pepsin at a final concentration of 0·64 per cent and hydrochloric acid at a concentration of 0·32 *N* (*p*H 1·5). The supernatant of the first treatment is referred to as the neutral extract and of the second as the acid peptic digest. The iron content was determined in each case, using the ortho-phenanthroline method described by Kaldor[4].

The importance of cooking hæmoglobin is seen from the results in Table 1, which also demonstrate the role of peptic digestion and the variability with which iron is released from different foods. Higher percentages are liberated from liver rather than from hæmoglobin, and the iron from the liver was shown to be derived more from the tissue iron (hæmosiderin and ferritin) rather than from the contained blood. The plant products yielded greater percentages than did hæmoglobin, but smaller absolute amounts per gram because of the lower total iron content. Phytic acid in silver beet did not prevent liberation of iron, but the amount of iron released was greater from old than from young leaves, and greater from stored than from fresh leaves.

These findings suggest a more rational basis for the compilation of dietary tables of iron values. However, the amount of iron released by peptic digestion of cooked food is not the only factor determining the amount of iron actually absorbed following ingestion of the food. The role of the other enzymes present in the gastro-intestinal tract has not been investigated. The needs of the body also have some regulating

TABLE 1. EXTRACTION OF IRON FROM FOODS

Food	Treatment		No of assays	Total iron content (mean)	Iron liberated (mean)	Per cent total iron liberated (mean)
Blood	Raw	saline extraction	2	524·4 μgm Fe/ml	5·8 μgm. Fe/ml.	1·1
		acid peptic digestion	6	518·9 ,,	12·5 ,,	2·4
	Cooked	saline extraction	5	535·7 ,,	8·0 ,,	1·5
		acid peptic digestion	15	529·6 ,,	53·5 ,,	10·1
Liver	Cooked	saline extraction	6	60·5 μgm. Fe/gm.	1·6 μgm. Fe/gm.	2·6
		acid peptic digestion	12	66·6 ,,	34·4 ,,	51·6
Plain flour	Cooked	water extraction	21	10·3 ,,	1·0 ,,	9·7
		acid peptic digestion	24	10·3 ,,	3·5 ,,	34·1
Silver beet	Cooked	saline extraction	6	18·3 ,,	3·0 ,,	16·5
		acid peptic digestion	36	20·5 ,,	13·2 ,,	64·4
Sultanas	Raw	saline extraction	9	17·5 ,,	0·8 ,,	4·7
		acid peptic digestion	10	18·4 ,,	8·6 ,,	46·9
	Cooked	saline extraction	11	14·8 ,,	2·4 ,,	16·1
		acid peptic digestion	9	17·3 ,,	10·7 ,,	62·1

effect on absorption but the mechanisms have not yet been elucidated.

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[1] Kaldor, I., *Aust. Ann Med.*, 6, 244 (1957).
[2] Brading, I, Kaldor, I, and George, E. P., *Aust. Ann. Med*, 6, 247 (1957).
[3] Moore, C V, and Dubach, R, in "Modern Trends in Blood Diseases" edit Wilkinson, J F, 109 (Butterworth, London, 1955)
[4] Kaldor, I, *Aust J Exp Biol Med. Sci.*, 31, 41 (1953).

Optical Rotation and Configuration of Poly-L-Tyrosine

IN a recent communication[1], Downie, Elliott and Hanby report that the optical rotatory dispersion of poly-L-tyrosine is unusual and, as a result, its configuration cannot be assigned by analogy with polypeptides having normal dispersion characteristics However, they interpret their data, together with unspecified infra-red spectral and X-ray diffraction observations of films, as indicating that "it is more probable that in dilute solutions in pyridine, in dimethyl formamide, and in aqueous alkali, poly-L-tyrosine is in a random coil configuration". Our study of this problem has led to somewhat different conclusions: that the configuration in pyridine and dimethyl formamide is that of the α-helix, whereas in alkaline solution the configuration is dependent on *p*H, indicative of the random-coil configuration. The basis of the configurational assignments will be published elsewhere in detail, but we wish to summarize them here and report the optical rotatory characteristics of the two forms.

Poly-L-tyrosine samples were prepared by modification of the Katchalski–Sela[2] procedure and shown to have molecular weights in the range 30,000–40,000 by combined sedimentation and viscosity measurements. In this molecular-weight range such measurements alone cannot establish configuration. Flow-birefringence measurements did, however, show that in dimethyl formamide/glycerol (3 : 1 by volume) the polymer exhibited strong positive birefringence. The same polymer sample when dissolved in aqueous alkali (*p*H 12·5, 1 *M* sodium chloride) – glycerol solution of equivalent viscosity failed to show any birefringence.

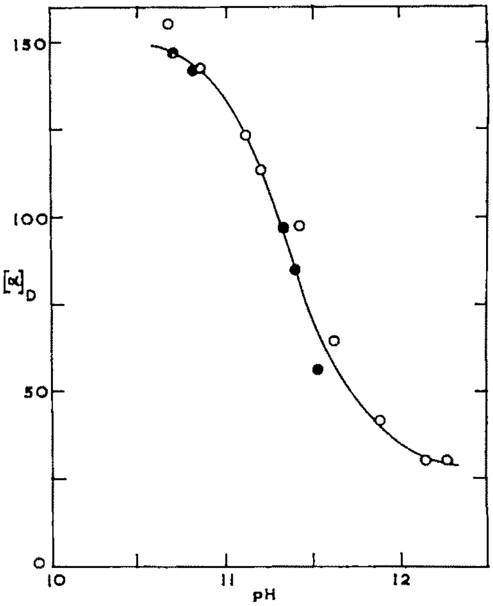

Fig. 1. Dependence on *p*H of specific optical rotation (at 589 mμ) of poly-L-tyrosine. All measurements were made in the 0·2 *M* sodium chloride. ● and ○ denote different polymer samples

The specific rotation of poly-L-tyrosine was measured in the *p*H range where it is soluble in aqueous solution, that is, above *p*H 10·6. The results, shown in Fig. 1, indicate a sharp fall in specific rotation coincident with the ionization of the tyrosine residue. The specific rotation of tyrosine itself changes only a few degrees in this same *p*H range. Since large changes in the optical rotation of poly-L-glutamic acid[3] and poly-L-lysine[4] were accompanied by a helix-coil transition, it was inferred that the same was true for poly-L-tyrosine. This was tested by measuring the sedimentation constant and specific viscosity as a function of *p*H at two different ionic strengths (0·2 and 1·0). Below *p*H 11·3, there was no dependence on ionic strength, indicating a rigid configuration. Above this *p*H the values were increasingly dependent on ionic strength, as would be expected for a flexible coil configuration. This supporting evidence leads to the conclusion that the configuration is helical near the solubility limit and randomly coiled beyond *p*H 12.

One can expect to observe the same helix-coil transition in organic solvents as the proportion of weak to strong hydrogen-bonding solvents is increased. This was found by measuring the specific rotation, $[\alpha]_D$, as a function of the concentration of dichloroacetic acid in dichloroacetic acid – dimethyl formamide mixtures[5]. The value of $[\alpha]_D$ fell sharply from 190° to 40° in the vicinity of 75 per cent dichloroacetic acid[6]. Since the strong birefringence indicated the helical form in dimethyl formamide, and since dichloroacetic acid has been shown to induce the solvated, random-coil configuration[7] and since, furthermore, the $[\alpha]_D$ values in these two solvents match the values of the two similarly assigned configurations in aqueous solution, it can be concluded that the α-helical configuration exists in dimethyl formamide, pyridine and at *p*H 10·7 in aqueous solution, while the random-coil configuration exists at high concentrations of dichloroacetic acid and above *p*H 11·8.

Having made this configurational assignment, it is of interest to examine the rotatory dispersion of the two forms in order to assess the magnitude of the perturbation produced in dispersion constants by the introduction of a strong chromophoric group close to the peptide bond chromophore. The relevant data for each configuration in two quite different solvents are listed in Table 1. The constant λ_0 was taken as 212 mμ in determining b_0 and the resulting plots were linear over the wave-length range investigated (450–700 mμ).

It is seen that each of the three constants has characteristic values for the two configurations. $[\alpha]_D$ for the helical form is more positive than the coil form by 120–150°. When data on this difference for six other, normally behaving, polypeptides is averaged and corrected to the residue weight of tyrosine, a predicted difference of only 50° is found.

Table 1. ROTATORY DISPERSION CONSTANTS OF POLY-L-TYROSINE

Solvent	$[\alpha]_D$	λ_0	b_0
0·15 *M* NaCl, *p*H 10·85	+ 150°	237 mμ	+ 540°
0·15 *M* NaCl, *p*H 11·20	+ 103	263	+ 520
0·15 *M* NaCl, *p*H 11·88	+ 38	316	+ 480
0·15 *M* NaCl, *p*H 12·27	+ 28	330	+ 450
Pyridine	+ 189	230	+ 560
Dimethyl formamide	+ 184	235	+ 550
Dichloroacetic acid			
,, with 20 per cent dimethyl formamide	+ 73	330	+ 470
,, with 4 per cent dimethyl formamide	+ 36	—	—

Thus, the sign of the difference for poly-L-tyrosine is normal, but the magnitude is about 2·5 times that normally observed.

The constant λ_0 usually has a value of about 210 mμ for the coil form, and increases to 300 mμ for the helix form. The behaviour found here is approximately the inverse.

The constant b_0 normally has a value of about −630 for the helical form and zero for the coil form. For poly-L-tyrosine we find instead positive values for both, and little dependence on configuration.

From this it is clear that the proximity of the tyrosine residue to the peptide bond greatly modifies the optical rotatory properties of both the helical and the coil forms. The close coupling between the two chromophores in either configuration, plus the fact that the primary helical arrangement of peptide bonds necessarily imposes a second helical arrangement on the chromophoric side-chains (tyrosine), introduces sufficient new pair-wise interactions to make the contribution of the original primary helix of peptide bonds to the optical rotatory properties essentially unrecognizable.

It might be conjectured that some of the changes in dispersion constants may be due to the helical form having the opposite screw sense to that normally occurring[7]. However, co-polymer studies to be described elsewhere rule out this possibility.

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[1] Downie, A. R., Elliott, A., and Hanby, W. E., *Nature*, **183**, 110 (1959).

[2] Katchalski, E., and Sela, M., *J. Amer. Chem. Soc.*, **75**, 5284 (1953).

[3] Doty, P., Wada, A., Yang, J. T., and Blout, E. R., *J. Polymer Sci.*, **23**, 851 (1957).

[4] Applequist, J., and Doty, P., Abstracts, American Chemical Society Meeting, San Francisco (April 7–11, 1958) (paper in preparation).

[5] Yang, J. T., and Doty, P., *J. Amer. Chem. Soc.*, **78**, 498 (1956).

[6] These data are shown in a discussion by Katchalski, E., Proc. Fourth International Congress of Biochemistry (Vienna), **9**, 28 (Pergamon Press, London, 1959). The b_0 values given in this discussion should be replaced by those stated here.

[7] Blout, E. R., and Karlson, R. H., *J. Amer. Chem. Soc.*, **80**, 1259 (1958).

A New Metabolite of Chloramphenicol

THE metabolism of chloramphenicol has been studied extensively in animals and in human subjects, with the recognition of the glucuronide as the principal metabolic product[1]. In addition, a small amount of the amino diol hydrolysis product and free chloramphenicol were found in the urine[1,2]. However, little attention has been given to the metabolism of this compound where deficiencies exist in the conjugating mechanism. The normal adult cat has been reported to be deficient in its capacity to form glucuronides with a number of synthetic substrates[3], and newborn animals generally have a low order of activity in the enzyme systems responsible for detoxication processes[4]. Consequently, the metabolism of chloramphenicol was investigated in the cat and in newborn human infants.

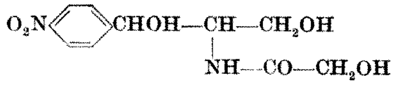
Following oral administration of chloramphenicol palmitate, the urine of adult cats and newborn infants was examined by paper chromatography. The solvent system was water-saturated butanol containing 2·5 per cent phenol and 2 per cent pyridine[1]. Strips of Whatman No. 1 paper were used in the descending method, and nitro compounds were detected by reduction with titanous chloride, diazotization and coupling as described elsewhere[1]. Aromatic nitro compounds were found in positions corresponding with chloramphenicol, the amino diol hydrolysis product of chloramphenicol, and the glucuronic acid conjugate. In addition, a fourth band located at R_F 0·70–0·75 was observed in cat urine and in the urine of newborn infants. A similar band was later observed in the urine of normal rats.

The presence of a glucuronide band in cat urine was of considerable interest in view of earlier reports concerning the absence of conjugation with phenolic compounds[3]. It is possible that conjugation of the terminal aliphatic alcohol group of chloramphenicol with glucuronic acid may occur through a different mechanism. The glucuronide nature of the product was confirmed by spraying the paper chromatograms with a solution of β-glucuronidase. After incubation for a few hours in a moist atmosphere, the strips were placed upon agar plates seeded with *Sarcina lutea*. Antimicrobial activity was found in the position corresponding with the chloramphenicol–glucuronide band, which showed no antimicrobial activity prior to hydrolysis.

The appearance of a new metabolite of chloramphenicol was also of interest because of its potential toxicity. This compound was isolated from cat urine, using a combination of methods involving adsorption on to charcoal, fractional elution with acetone–water mixtures, 24-plate counter-current extraction with a two-phase solvent system consisting of 270 ml. water, 30 ml. *iso*propanol and 300 ml. ethyl acetate, and by paper chromatography. The final crystalline product had an off-white colour and gave a melting point of 155–156° C. (uncorr.). It showed 4 per cent of the antimicrobial activity of chloramphenicol against *Shigella sonnei* in turbidimetric assays. $E(1\%, 1\text{ cm.}) = 367$ at 274 mμ (ethanol).

Analysis: Calc. for $C_{11}H_{14}N_2O_6$ (270·24): C, 48·89; H, 5·22; N, 10·37. Found: C, 49·26, 49·34; H, 5·38, 5·34; N, 10·44.

The analytical results are in good agreement with the structure for D(−)*threo*-1-*p*-nitrophenyl-2-hydroxyacetamido-1,3-propanediol:



$$O_2N{-}C_6H_4{-}CHOH{-}\underset{\displaystyle NH{-}CO{-}CH_2OH}{\underset{|}{CH}}{-}CH_2OH$$

The metabolite is a glycolic acid amide analogue of chloramphenicol, apparently resulting from dehalogenation and reduction of the dichloracetate group in the parent compound. This analogue was prepared by synthesis, using the acid chloride procedure described by Rebstock[5]. The product showed the same melting point and mixed melting point as the metabolite from cat urine. $E(1\%, 1 \text{ cm.}) = 375$ at 274 mμ (ethanol). The infra-red absorbance spectra of the two preparations were identical, and they showed the same solubility characteristics in the counter-current extraction system already described, as well as having the same R_F values in paper chromatograms, and the same antimicrobial activity. The metabolite present in the urine of newborn human infants was also isolated by the same procedure, and proved to be identical with the product from cat urine in its counter-current distribution, paper chromatography, and antimicrobial activity.

Preliminary acute toxicity data obtained by intravenous administration of the synthetic product gave an $LD50$ value of $2{,}950 \pm 81$ mgm./kgm. in rats, and $2{,}950 \pm 72$ mgm./kgm. in mice, indicating that the metabolic product has a very low order of toxicity.

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[1] Glazko, A. J., Dill, W. A., and Rebstock, M. C., *J. Biol. Chem.*, **183**, 679 (1950).

[2] Glazko, A. J., Wolf, L. M., Dill, W. A., and Bratton, A. C., *J. Pharm. Exp. Therap.*, **96**, 445 (1949).

[3] Robinson, D., and Williams, R. T., *Biochem. J.*, **86**, 23, *P* (1958).

[4] Driscoll, S. G., and Hsia, D. Y-Y., *Pediatrics* (Supp.), **22**, 785 (1958).

[5] Rebstock, M. C., *J. Amer. Chem. Soc.*, **72**, 4800 (1950).

Effect of Glycosylation on the Enzymic Oxidation and Translocation of Flavonoids

There is a possibility that a planar structure may be one of the factors which determines the ability of a flavonoid to function as a substrate for plant polyphenoloxidases. It is also possible that a planar structure may adversely affect the ease of translocation of flavonoids in vegetable tissues.

As a result of an extensive study of the paper chromatographic behaviour of phenolic substances it has been established that all flavonoids with a planar structure have zero R_F in water, or in aqueous solvents containing up to 2 per cent of acid[1]. The molecular model in Fig. 1 illustrates the planar structure of quercetin. Other polynuclear phenols with 'flat' molecules, for example, ellagic acid, resveratrol and galloflavin, have also been shown to have zero R_F values in water. It is known that cellulose absorbs phenolic substances, and it appears that when a molecule contains two phenolic rings in the same plane the adsorption by cellulose is sufficient to prevent elution of the adsorbed substance by aqueous solvents.

The R_F values in aqueous solvents of substances such as gallic and caffeic acids are significantly increased by combination with glucose and quinic acid.

	R_F in 2 per cent acetic acid
Gallic acid	0·44
Glucogallin	0·64
Caffeic acid	0·25
Chlorogenic acid	0·56

As the R_F values in water of flavonol-3-glycosides are significantly greater than zero[2], it was originally assumed that the increased R_F was due to combination with water-soluble sugar residues. However, it has been found that both kaempferol-4′-arabinoside and quercetin-4′-glucoside have zero R_F values, and Geissmann has also recorded zero R_F in water for quercetin-7-glucoside (quercimeritrin)[3].

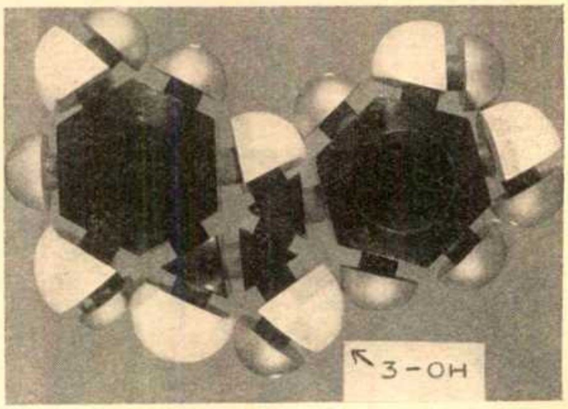

Fig. 1. Planar structure of the quercetin molecule built up from Courtauld's atomic models. Note that the hydrogen atom of the 3-hydroxyl group can only just be accommodated in the planar structure

The introduction of a sugar residue into the 4′ or 7 positions of a flavonol does not destroy the planarity of the molecule. On the other hand, as will be apparent from an inspection of the molecular model of quercetin, substitution of a large group in the 3-position cannot be achieved without a loss in planarity. Glycosylation only increases R_F values in aqueous solvents if in the 3-position, so that it appears that the increase is a consequence of loss of planarity and not to combination with a large water-soluble group.

It was shown by Baruah and Swain that potato polyphenoloxidase catalysed the oxidation of quercetin, myricetin and luteolin, but that the 3-glucosides of quercetin and myricetin were not oxidized[4]. This confirmed and extended earlier work by Roberts and Wood[5]. These results suggest that plant oxidases act preferentially upon flat molecules, a planar structure facilitating adsorption by the active enzyme surface. One might therefore anticipate that glycosides of luteolin, quercetin and myricetin might function as substrates so long as the sugar group was not in the 3-position, and it has been shown by Baruah and Swain[4] that the 7-diglucoside of luteolin (chærophyllin) undergoes oxidation in the presence of the potato oxidase. However, it was also stated that quercimeritrin was not oxidized, and this is inconsistent with the hypothesis advanced.

Samples of these two flavonoids were made available by Dr. Swain, and, as expected, it was found that chærophyllin had zero R_F in 2 per cent acetic acid. However, the substance labelled quercimeritrin was almost certainly inaccurately described, for it was resolved by two-way paper chromatography into at least five components, only one of which had zero R_F in aqueous solvents. It has been confirmed in this laboratory that an authentic specimen of quercimeritrin has an R_F of zero in 2 per cent acetic acid, and it may be safely concluded that Baruah and Swain's findings for quercimeritrin are incorrect. In summarizing the relation between substrate activity and R_F values in aqueous solvents (Table 1), results for quercimeritrin have not therefore been included.

The strong adsorption of flavones and flavonol aglycones by cellulose may well restrict their mobility

Table 1. SUBSTRATE ACTIVITIES AND R_F VALUES IN AQUEOUS SOLVENTS OF FLAVONOIDS

Substrate	Structure	R_F value	Oxidized by enzyme
Quercetin	Planar	0·00	Yes*
Quercetin-3-rhamnoside	Non-planar	0·19	No
Quercetin-3-rhamno-glucoside	Non-planar	0·29	No
Myricetin	Planar	0·00	Yes
Myricetin-3-rhamnoside	Non-planar	0·23	No
Luteolin	Planar	0·00	Yes
Luteolin-7-diglucoside	Planar	0·00	Yes

* Roberts and Wood (ref. 5) stated that quercetin was not oxidized by the tea oxidase. This was an inaccurate observation due to the very low solubility in water of quercetin.

in plant tissues, and it is suggested that glycosylation in the 3-position is necessary for translocation to take place. A paradoxical situation may now arise, for the planarity of the molecule, which is necessary for any physiological activity dependent upon enzymic oxidation, will hinder translocation. On the other hand, the more mobile glycoside may be physiologically inactive unless there is a glycosidase present to regenerate the active aglycone. Considerations such as these may also be relevant to the activity of certain flavonoids on vascular resistance or capillary permeability in animals[6,7].

I wish to thank Dr. T. Swain for helpful discussions, and Dr. D. E. Hathway and Profs. R. D. Haworth and L. Hörhammer for specimens of resveratrol, galloflavin and flavonol-4′-glycosides. Thanks are also due to the Indian Tea Association for permission to publish this communication.

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[1] Roberts, E. A. H., Symp. Soc. Leather Trades' Chemists, 87 (Cambridge, 1956).

[2] Roberts, E. A. H., Cartwright, R. A., and Wood, D. J., *J. Sci. Food Agric.*, **7**, 637 (1956).

[3] Geissman, T. A., in "Modern Methods of Plant Analysis", by Paech, K., and Tracey, M. V., **3** (Berlin, Göttingen, Heidelberg: Springer-Verlag, 1955).

[4] Baruah, P., and Swain, T., *J. Sci. Food Agric.*, **10**, 125 (1959).

[5] Roberts, E. A. H., and Wood, D. J., *Nature*, **167**, 608 (1951).

[6] Lockett, M. F., "The Pharmacology of Plant Phenolics", 81 (Academic Press, London, 1959).

[7] Lavollay, J., and Neumann, J., "The Pharmacology of Plant Phenolics", 103 (Academic Press, London, 1959).

Need for Inorganic Phosphate in Oxidations stimulated by Dinitrophenol

Recently we have reported, in disagreement with the widely accepted view, that the maximal rate of oxygen uptake by respiring rat-liver or rat-heart mitochondria with glutamate as substrate is not obtained with 2,4-dinitrophenol unless phosphate is also present[1]. With succinate as substrate, on the other hand, the same initial rate of oxygen uptake was found with dinitrophenol as with inorganic phosphate. We concluded that a link in the respiratory chain between glutamate and cytochrome *b* required phosphate even in the presence of dinitrophenol, a conclusion which seemed to have important implications for theories of the mechanism of the uncoupling of oxidative phosphorylation by dinitrophenol.

We now wish to report that further studies have provided evidence that dinitrophenol can replace inorganic phosphate in the respiratory chain. There is, therefore, no need to modify our earlier view[2] that dinitrophenol reacts with an energy-rich intermediate in oxidative phosphorylation formed prior to the intervention of inorganic phosphate.

The new evidence we have obtained is:

(1) Substrates, such as malate and β-hydroxybutyrate, which operate through nicotinamide–adenine dinucleotides, are oxidized at the same rate in the presence of 2,4-dinitrophenol alone as with dinitrophenol and inorganic phosphate together.

(2) The results with glutamate are explained by the finding that a large part of the measured oxygen uptake with this substrate is not concerned with the oxidation of glutamate to α-ketoglutarate, but with the further oxidation of the α-ketoglutarate. This is shown by the fact that the α-ketoglutarate (μmoles) found in the presence of phosphate was less than 1 per cent of the oxygen uptake (μatoms). Malonate (0·02 *M*) inhibited by 70 per cent the rate of oxygen uptake with liver mitochondria. The requirement for inorganic phosphate for maximal rates of oxygen uptake is explained by the fact that dinitrophenol cannot replace inorganic phosphate in the 'substrate-linked' phosphorylation step associated with α-ketoglutarate oxidation[3].

The rapidity with which the α-ketoglutarate formed from glutamate is oxidized by mitochondrial preparations was unexpected. It is clear that in the absence of an inhibitor of α-ketoglutarate oxidation, glutamate is not a suitable substrate for studying oxidative phosphorylation and respiratory control associated with the oxidation of reduced nicotinamide–adenine dinucleotides.

We wish to thank Prof. F. Lynen for a stimulating discussion which led us to examine more closely the products of oxidation of glutamate. This work was supported by a grant from the Netherlands Organization for National Health Research T.N.O. and the Koningin Wilhelmina Fund for Cancer Research.

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[1] Borst, P., and Slater, E. C., *Nature*, **184**, 1396 (1959).

[2] Slater, E. C., *Nature*, **172**, 975 (1953).

[3] Judah, J. D., *Biochem. J.*, **49**, 271 (1951). Hunter, F. E., in "Phosphorus Metabolism", **1**, 297 (The Johns Hopkins Press, Baltimore, 1959).

PHARMACOLOGY

Histamine Sensitivity in Children after *Pertussis* Infection

IT has been shown in recent years that mice and rats are rendered hypersensitive to the action of histamine, 5-hydroxytryptamine and anaphylactic sensitization by pre-treatment with *Hæmophilus pertussis* vaccine[1-4]. These properties are present only in organisms in the smooth phase and are not shared by other members of the *Hæmophilus* family[5,6]. The sensitizing action is not seen in the guinea pig and the rabbit; on the other hand, there is some evidence that these two species are rendered slightly more resistant to histamine after such treatment[7]. Rats and mice are relatively resistant to histamine and anaphylactic shock, whereas guinea pigs and rabbits are susceptible. It was therefore of interest to study whether the reaction in man follows a similar pattern to the former two species or the latter. Recently there was an epidemic of whooping cough in Darbhanga and advantage was taken of this for such a study.

Histamine sensitivity was tested by the method developed by Jha in this laboratory. Briefly, it consists of placing on the fore-arm a lint soaked in an aqueous solution of histamine acid phosphate (10^{-5} conc. as base) and another in *N* saline. The anode of a 6-V. dry battery is placed on the histamine lint and the cathode on the other and current is allowed to pass for exactly 2 min. The reaction is read in 15 min. time. The normal reaction consists of erythema and occasional papules which never cover more than a quarter of the area of application, and is arbitrarily classified as (+). When papules cover half the area the reaction is read as (++) and when more than three-quarters are involved as (+++). A wheal formation is indicative of a marked hypersensitivity and is designated as (++++). By giving a score 2 to each (+), the average reaction can be expressed as a percentage of the maximal reaction possible. In this system of scoring, normal reaction is between 0 and 20 per cent, and values above 50 per cent represent hypersensitivity.

Sensitivity to 5-hydroxytryptamine, and the histamine liberator compound 48/80 can also be studied similarly.

Thirty children in the typical stage of 'whoop' in the age group 1–10 years were selected for the study, and the sensitivity to histamine, 5-hydroxytryptamine and compound 48/80 was determined separately. It was found that nearly all the patients were maximally sensitive to histamine (average reaction 88 per cent), and slightly sensitive to 5-hydroxytryptamine (average reaction 40 per cent). Wheals developed after histamine iontophoresis in majority of the patients, but were never encountered after 5-hydroxytryptamine. The patients failed to exhibit sensitivity to compound 48/80 (average reaction less than 10 per cent).

Thus it is seen that infection with *Hœmophilus pertussis* produces a condition in children similar to that noted in rats and mice. It is difficult to say at present whether such children are more susceptible to allergic reactions as well.

It has been reported that the anaphylactoid reaction (œdema) in rats is mediated through 5-hydroxytryptamine[8]. Such a conclusion may not be true for man, for although histamine in many cases produced an urticarial wheal, 5-hydroxytryptamine failed to do so when applied in a similar concentration. It may be noted that 5-hydroxytryptamine is 200 times as potent as histamine in producing the œdema reaction in the rat[9].

The fact that, even in patients maximally sensitive to histamine, compound 48/80 failed to produce significant reaction leads to the indirect inference that histamine content of the skin of children is low, particularly as when the experiments were repeated in adults, in many cases, typical reactions were obtained.

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[1] Parfentjev and Goodline, *J. Pharmacol.*, **92**, 411 (1948).
[2] Malkiel and Hargis, *Proc. Soc. Exp. Biol.*, **80**, 122 (1952).
[3] Sanyal and West, *J. Physiol.*, **142**, 571 (1958).
[4] Kallos and Kallos-Deffner, *Int. Arch. Allergy*, **11**, 237 (1957).
[5] Kind, *J. Immunol.*, **70**, 411 (1953).
[6] Maitland, Kohn and MacDonald, *J. Hyg.*, **53**, 196 (1955).
[7] Stronk and Pittman, *J. Infect. Dis.*, **96**, 152 (1955).
[8] Parratt and West, *J. Physiol.*, **135**, 10,*P* (1956).
[9] Parratt, Ph.D. thesis, University of London (1958).

Interactions between Pharmacodynamic and Placebo Effects in Drug Evaluations in Man

IN the course of the development of a method sufficiently sensitive to evaluate the effects of small doses of tranquillizers in man, evidence of interactions between pharmacodynamic and placebo effects of drugs was adduced. Since placebo action (that is, the response to the act of taking medication as distinguished from the pharmacodynamic action of the medicament) must be taken into account in analysing human reactions to drugs, we feel that our findings have implications of general interest as well as of practical importance in designing methods for observing and evaluating the effects of drugs in man.

The influence of competitive mental work (a variant of the Stroop test[1]) on high-amplitude finger tremor and its modification by sedative drugs was studied in 12 male medical students. To control bias as well as placebo effects, dummy medication[2] in a capsule of identical appearance was administered in a classic double-blind arrangement[3]. To evaluate the extent of the placebo influence on the results, the test was also conducted with neither drug nor dummy. The tests were randomized.

The pertinent findings may be briefly summarized. After dummy medication had been given, the rate of high-amplitude finger tremor rose during the test from an average of 60 to 198 per min.; after 100 mgm. of pentobarbital sodium, it rose to 162; when neither drug nor dummy had been given, it rose to only 152 per min. By the sign test, statistical analysis indicated that each of the latter two is significantly smaller than the rate after the dummy capsule ($P < 0{\cdot}05$). However, the rate after 50 mgm. of pentobarbital did not differ significantly from that after the dummy.

Depending on the control used, the data may provide two diametrically opposed answers to the question of the effect of a sedative on high-amplitude tremor after competitive mental work. Thus, if the rate of tremor when neither drug nor dummy had been given is used as a control, 100 mgm. of pentobarbital is seen to have increased the tremor-rate. However, when the effect of 100 mgm. of pentobarbital is compared with the tremor-rate after a dummy, the drug appears to have decreased the tremor-rate. These results indicate that, with reference to the untreated, the drug made matters worse, whereas with reference to a placebo, the drug made matters better.

The increase in tremor-rate produced by 50 mgm. of pentobarbital is seen to fall within the range of the placebo effect. Consequently, the extent of the pharmacodynamic effect, if any, is uncertain. As a corollary, the placebo effect of the active agent is also uncertain, but it does confirm the stressful effect of the act of medicating.

The implications of these results are important for the clinical evaluation of drugs. The findings indicate that it is not always possible to anticipate the nature or the direction of placebo reaction in a particular circumstance. In this work, after it had been determined that the competitive mental work test did regularly and predictably increase the rate of high-amplitude finger tremor, it was presumed that the dummy would act as a placebo, that is, it would reduce the rate of high-amplitude finger tremor during the competitive mental work. However, it was found that the dummy medication intensified high-amplitude finger tremor and, as a result, tremor-rate after a sedative was also greater than when no treatment was given.

It is obvious from these trials that difficulty may arise in defining the base-line for the estimation of pharmacodynamic actions in man, hence in making absolute measurements of drug action. In this study, because drug and placebo effects were opposed, drug action could be measured in terms of the extent by which the drug diminished the placebo reaction. In the situation in which drug effect is in the same direction, but exceeds placebo effects, the question of how to obtain an absolute measure of drug effect is a difficult one, for the results may not indicate whether the drug action supplements placebo effect or accompanies it. In other cases, in which the subtraction indicates no effect in excess of placebo effect, it does not rule out the possibility of the complete masking of drug effect. This also brings up the question of the sensitivity of the method requisite for drug evaluations since, as the relative proportions of placebo and pharmacodynamic actions in the total effect change, the ability of a method to discriminate between them, hence its sensitivity, also changes[4].

It is of especial interest, therefore, that a so-called 'negative' placebo effect is not necessarily an obstacle to the development of a useful and sensitive method of drug evaluation, for if the nature and the direction of placebo reaction, hence the base-line, can be identified, drug effect can be measured accurately enough; indeed, sometimes more accurately when they are opposed than when the effect of one obscures the effect of the other, as it may, when they develop simultaneously and in the same direction.

In the present instance, the dummy induced, not a typical placebo effect but, instead, a significant and useful stress, and the competitive mental work test merely provided an additional challenge, while the pharmacodynamic effect opposed both. It can be assumed that, regardless of how well they were re-assured that there was no hazard, the use of volunteer medical students[5] who knew that they were participating in a drug investigation led to the unexpected and uniform placebo response. It is possible that, were the subjects entirely unaware of participation in an investigation and, more specifically, of drug testing, both placebo and pharmacodynamic actions might have been in the same, instead of opposite, directions. However this may be, this circumstance may nevertheless provide a well-controlled, sensitized setting for the evaluation of the small doses of tranquillizers so commonly used in general practice.

The importance of 'negative' placebo effects in simulating toxic effects of drugs has been stressed in the literature[6]. In the present case, such an effect has been measured and has been shown to be regular and predictable. Because of the several varieties of placebo reactions as well as their potential for interaction with pharmacodynamic effects of drugs[7], this dynamic quality, which is inseparable from the act of drug administration in man, must be taken into account, and controls must be used which identify the direction as well as the magnitude of placebo effects. For this it may be necessary to use the unblinded 'no-drug-at-all' control to complement the usual identical dummies in the double-blind setting.

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[1] Stroop, J. R., *J. Exp. Psych.*, **18**, 643 (1935).
[2] Gaddum, J. H., *Proc. Roy. Soc. Med.*, **47**, 195 (1954).
[3] Gold, H., *Amer. J. Med.*, **17**, 722 (1954).
[4] Modell, W., and Houde, R. W., *J. Amer. Med. Assoc.*, **167**, 2190 (1958).
[5] Lasagna, L., and von Felsinger, J. M., *Science*, **120**, 359 (1954).
[6] Wolf, S., and Pinsky, R. H., *J. Amer. Med. Assoc.*, **155**, 339 (1954).
[7] Wolff, H. G., and Goodell, H., *Proc. Assoc. Res. Nerv. Ment. Dis.*, **23**, 434 (1943).

ANIMAL PHYSIOLOGY

Relation of Eye Dominance and Eye Closure

It is a well-known fact that while our vision is binocular, one eye (more often the right than the left) is in fact preferred or dominant in fixation and sighting. Much has been published on this subject, especially in its relation to handedness. We conjectured that if one eye is dominant in sighting, the other, that is, the non-dominant eye, would logically be expected to close more easily in order to enhance the use of the dominant eye, and as a corollary, the dominant eye would be more resistant to closure unilaterally because of the very nature of its dominance. In putting this theory to test, we used the students of our senior and junior anatomy classes (total 145, ranging from 18 to 24) as subjects. Eye dominance was first ascertained by three or more trials of the cone test in each case, and then the subjects were asked to close the right and left eyes singly and alternately for a number of times and to say which eye they found could close with less effort. This latter test was subjective; in some cases, however, we were able to see quite plainly for ourselves. The results of our investigation are given in Table 1.

Table 1

No. of subjects	Dominant eye	Ease in closing
63	*R*	*L*
31	*L*	*R*
29	*R*	*R*
16	*L*	*L*
4	*R*	*ND*
2	*L*	*ND*

L, Left eye; *R*, right eye; *ND*, no difference between the eyes.

The above results show that two-thirds of the subjects could close the non-dominant eye more easily than the dominant eye, while one-third the reverse. The value of chi-square obtained ($\chi^2 = 15\cdot527$ and $P = 0\cdot01$) shows a significant correlation between eye dominance and the relative ease of closure of the non-dominant eye. In addition, the results show that the differential function of the orbicularis oculi, innervated by the facial nerve, is a component in the dynamic complex of eye dominance.

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Presence of Aldosterone-stimulating Hormone in Urine

THE secretion of aldosterone differs from that of other steroids of the adrenal cortex in that it is not directly under pituitary control. Current concepts favour a location in the diencephalon for the centre which controls aldosterone secretion[1]. The transmission of the stimulus from the centre to the effector gland cannot be nervous, since the isolated gland can respond to suitable provocations by an increased secretion of aldosterone[2]. It is therefore concluded that the stimulus is humoral.

Evidence for the presence in the urine of adrenalectomized rats deprived of sodium of a substance which modifies the secretion of an electrolyte-active steroid when injected into intact rats has been presented by Orti *et al.*[3]. This substance may be the hypothetical hormone responsible for the stimulation of aldosterone synthesis or release. The present investigation confirms the presence of this substance in urine under certain conditions, and evidence is presented that it is an 'aldosterone-stimulating hormone'.

Rats of Wistar strain, weighing about 150 gm., were bilaterally adrenalectomized and maintained on rat cake, with 1 per cent sodium chloride solution to drink. Groups of 20 of these animals were deprived of food and drink for 36 hr., and their urine collected over the last 12 hr. of this period. This urine was injected subcutaneously into groups of 5 intact rats, together with a sodium load of 1 ml. of 0·45 per cent sodium chloride solution per 50 gm. body-weight, administered intraperitoneally. 5 intact rats served as controls, receiving a similar sodium load intraperitoneally and a volume of urine from adrenalectomized rats receiving 1 per cent saline was given subcutaneously, equal in volume to that of the urine injected into the experimental group. Urine was collected individually from the 10 rats at intervals of 40 and 80 min. after the injection of the urine. Cross-over tests were made three days later. Statistical analysis of variance shows that the administration of urine from the salt-deprived animals resulted in a lowering of the sodium/potassium ratio in the urine of the intact animals compared with the control group, as shown in Table 1.

Table 1

No. of rats	Time (min.)	Mean sodium/potassium, experimental	Mean sodium/potassium, control	*P*
40	40	1·71	2·34	0·005
40	80	1·09	1·37	0·01

The urine from the experimental and control groups was collected in separate pools. This urine was hydrolysed for 48 hr., extracted with chloroform and run on the chromatographic system of Zaffaroni *et al.*[4] (toluene–propyleneglycol), or the '*B5*' system of Bush[5]. Insufficient material was present for a definite chemical identification of aldosterone to be made. The material running in the position of aldosterone was eluted from paper and bio-assayed in adrenalectomized rats. The urine from the experimental group possessed sodium-retaining activity.

Serial assay on a similar pool of urine showed that the substance was stable at 4° C. for seven days. It would also withstand boiling for 5 min. with little loss of activity.

These observations substantiate the presence in the urine of dehydrated adrenalectomized rats of a substance which, when injected into intact rats, causes a lowering of urinary sodium/potassium ratio, and circumstantial evidence is presented that this is due to the release of aldosterone. Corticotrophin, 1 U.S.P. unit per rat, did not possess this activity. It is concluded that the substance responsible for this effect is 'aldosterone-stimulating hormone'.

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[1] Farrell, G. L., "Recent Prog. Hormone Research", **15**, 275 (1959).
[2] Flemming, R., and Farrell, G., *Endocrinol.*, **59**, 360 (1956).
[3] Orti, E., Ralli, E. P., Laken, B., and Dumm, M. E., *Amer. J. Physiol.*, **191**, 323 (1957).
[4] Zaffaroni, A., Burton, R. B., and Keutman, E. H., *Science*, **111**, 6 (1950).
[5] Bush, I. E., *Biochem. J.*, **50**, 370 (1952).

Biological Effects of Aromatic Extracts from Urine of Schizophrenics

Wada and Gibson[1] have recently reported that aromatic material extracted from the urine of schizophrenic patients induces behavioural and electroencephalographic abnormalities when injected intracisternally and intraventricularly in cats and monkeys. They used the fraction extracted by eluting with phenol-saturated water the material adsorbed on activated charcoal, according to the method of McGeer *et al.*[2].

Similar observations were made in the course of a series of experiments including the study of biological effects produced in mice, rabbits and dogs by aromatic fractions extracted from the urines of normal and schizophrenic subjects. The extracts were obtained by Decker's method[3], which is based upon elution with a water–methanol–butanol–ammonia mixture of the material adsorbed on activated charcoal. Of the group of substances thus extracted only the methanol-soluble fraction was investigated. Previous paper chromatographic analysis[4] of this fraction has shown that a larger number of aromatic substances is present in the urine of schizophrenic subjects.

The biological studies of such extracts included the determination of (*a*) the acute toxicity following intraperitoneal injection in mice; (*b*) the chronic toxicity in mice with histological examination of the brain and other viscera; (*c*) the behavioural effects following intracerebral injections in the rabbit; (*d*) the effect on the electrical activity of the cerebral cortex following topical applications in rabbits and dogs.

Both the aromatic fraction extracted from the urine of schizophrenic subjects and that from urine of normals have toxic properties. However, the fraction from schizophrenic patients appears to possess a higher acute toxicity and produces at lower dose-levels, upon chronic administration, degenerative changes of nerve cells of brain-stem nuclei. Furthermore, the aromatic urinary fraction from schizophrenic subjects has definite convulsant properties but only when brought into direct contact with the central nervous system. Intracerebral injections of minute amounts of the diluted extracts are readily followed by prolonged and generalized convulsions.

Direct applications of the diluted material to very small areas (2 sq. mm.) of the surface of the cerebral cortex cause localized convulsive alterations of the electrocorticogram.

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[1] Wada, J., and Gibson, W. C., *Amer. Med. Soc., Arch. Neurol. Psychiat.*, **81**, 747 (1959).

[2] McGeer, E. G., Brown, W. T., and McGeer, P. L., *J. Nerv. Ment. Diseases*, **125**. 176 (1957).

[3] Decker, P., *Z. ges. inn. Med. u. ihre Grenzgeb.*, **9**, 966 (1954).

[4] Kemali, D., and Buscaino, V. M., "Chemical Concepts of Psychosis", ed. by Rinkel, M., 219 (McDowell-Obolensky, New York, 1958).

Proximo-Distal Movement along the Axon of Protein synthesized in the Perikaryon of Regenerating Neurons

DURING regeneration of crushed nerve-fibres of spinal ganglion cells, the turnover rate of proteins in the ganglia (*C*.7 and *C*.8) increases extensively, as compared with the controls[1]. This finding, on one hand, justifies the increase in protein in the perikaryon of the regenerating neuron[1,2]; on the other hand, it supports Weiss's opinion[3] that some of the new proteins leave the body-cell and reach the growing end of the axon.

Movement of the protein along the axon can be studied by tagged compounds[4,5], especially when a set experimental condition is followed; that is, *in vivo* only the perikaryon of regenerating nerve cells must synthesize labelled proteins from tagged precursors (for example, *C*14-amino-acids). Thus, if the 'new' proteins move along the axon, it should be possible to collect them at the regenerating end of the nerve fibre. It is clear that a labelled precursor of proteins in the blood would contaminate all boundaries of crushed neurons, giving rise to difficulties in the evaluation of the phenomenon.

Hypoglossal and vagus nerves of adult rabbits were exposed on one side and crushed by forceps cooled to $-70°$ C.; the former at the level of the intermediate tendon of the digastric muscle; the latter at the carotid sinus. After 13 days, an opening was made on the middle nuchal line and a small window ($\sim$ 3 mm. diam.) was made through the atlo-occipital membrane. After removing the tela chorioidea of the fourth ventricle and drying the cerebrospinal fluid by filter paper, when necessary, the calamus scriptorius becomes clearly visible. About 0·001 ml. of Krebs and Henseleit solution[6], containing 0·2 gm. per cent of glucose, 5·55 mgm. per cent of L-threonine-^{14}C (*S.A.*, 32·9 μc./mgm.) and 7·8 mgm. per cent of L-valine-^{14}C (*S.A.*, 68·5 μc./mgm.), was placed on the calamus, using a micrometer syringe; the procedure was repeated every 20 min. for 5 hr. In this way it was possible to concentrate the tagged amino-acids in the region of the hypoglossal nucleus and dorsal nucleus of the vagus, without introducing significant quantities of carbon-14 compounds into the blood or cerebrospinal fluid.

At different intervals after the experiment, the growing segments of the XII and X nerves, as well as the homologous segments of the other side, used as controls, were removed. Each tissue sample was homogenized in 9 vol. of 5 per cent cold trichloracetic acid and the suspension centrifuged; the procedure was repeated twice and the residue was washed successively three times with acetone and three times with ether. Finally, the dry residue was hydrolysed in 1 ml. of 6 *N* hydrochloric acid in a sealed tube at 115° C. for 18 hr. The hydrolysate was brought to dryness at 50° C. in air stream, taken up in water and transferred to a planchet for determination of radioactivity in a gas-flow counter.

The results can be summarized as follows: (1) During the first 30 hr. after commencement of the experiment, the regenerating segments of the XII and X nerves showed no radioactivity. (2) In the next 10 hr., the regenerating segments displayed a significant amount of radioactivity. This radioactivity was inseparable from threonine and valine, as was demonstrated by autoradiograms of the amino-acids from the protein hydrolysate separated by paper chromatography. The control segments did not show any radioactivity. (3) Between the fortieth and seventy-second hour, radioactivity was still present within the regenerating segments; no radioactivity was found in the homologous normal segments.

A detailed account of the experiments will be published elsewhere.

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[1] Miani, N., Rizzoli, A., and Bucciante, G., *J. Neurochem.* (in the press).

[2] Brattgård, S.-O., Edström, J.-E., and Hydén, H., *J. Neurochem.*, **1**, 316 (1957).

[3] Weiss, P., and Hiscoe, H. B., *J. Exp. Zool.*, **107**, 315 (1948).

[4] Gerard, R. W., in Weiss, P., "Genetic Neurolog." (Univ. of Chicago Press, Chicago, 1950).

[5] Samuels, A. J., Boyarrsky, L. L., Gerard, R. W., Libet, B., and Brust, M., *Amer. J. Physiol.*, **164**, 1 (1951).

[6] Krebs, H. A., and Henseleit, K., *Hoppe-Seyl. Z. physiol. Chem.*, **210**, 33 (1932).

Hæmostatic Mechanisms in the Animal Arterial Wall

DEPOSITION of fibrin on the interior surfaces of arteries after intimal injury forms the basis of the thrombogenic theory of the pathogenesis of arteriosclerosis[1]. Such fibrin deposits, covered by endothelium, have repeatedly been observed[2–6].

Fibrin deposition in the organism is determined by a hæmostatic balance between fibrin formation and fibrin resolution, which is regulated by the mechanisms of blood coagulation and fibrinolysis[7,8]. In order to try to substantiate the role of fibrin deposition as a primary process in the development of arteriosclerotic lesions, we studied the thromboplastic and fibrinolytic activities of the separated layers of the wall of the human aorta. We found high thromboplastic activity in the central layers (intima and media) and less activity in the adventitia. Little or no fibrinolytic activity was present in intima or media, while adventitia was highly fibrinolytic[9]. These results were verified by additional studies comprising samples of aortæ with varying degrees of atheromatosis[10]. The high thromboplastic activity of the human aorta (including intima) was recently confirmed[11].

It has proved difficult, if not impossible, to produce experimentally in animals pathological changes identical with human arteriosclerotic lesions. This observed fact has never been satisfactorily explained.

However, an investigation of the fibrinolytic activity of samples of aortæ from monkeys demonstrated considerable activity in intima and media[10]. Furthermore, in bovine aorta low thromboplastic activity was found in the intima, while media and adventitia had higher activity[12]. Hence, animal arteries apparently differ from human vessels in their hæmostatic systems.

In order to elucidate this problem we have recently compared the aortic walls in some animal species. The procedures were the same as applied in our previous studies[10]. Freshly drawn citrated rabbit plasma served as test substrate for the thromboplastic activity. The concentration of plasminogen activator in the tissue was estimated after extraction with 2 *M* potassium thiocyanate[13]. The results of this investigation are summarized in Table 1.

Table 1. ANIMAL SPECIES ARRANGED ACCORDING TO HÆMOSTATIC SYSTEM IN ARTERIAL INTIMA

Fibrinolytic activity in decreasing order; thromboplastic activity in increasing order. The results represent average values

Fibrinolytic activity

Animal	Adv.	M.		I.
Monkey	+ + +	+		+
Horse	+ + +	+		(+)
Dog	+ +	(+)		(+)
Rat	+ +		+ +	
Guinea pig	+		(+)	
Pig	+ + +	0		0
Ox	+	0		0
Rabbit	0	0		0

Thromboplastic activity

Animal	Adv.	M.		I.
Rat	Inhib.		Inhib.	
Dog	0	(+)		0
Guinea pig	+		(+)	
Monkey	+	+		(+)
Ox	+	+ + +		+
Pig	+ + +	+ +		+
Horse	+ +	+ + +		+ +
Rabbit	(0)	+ +		+ + +

Adv., adventitia; M., media; I., intima; Inhib., inhibition.

It is apparent that great variations in the hæmostatic systems in different animal species exist. It is interesting to note that the rabbit, a preferred animal for experiments on connective tissue and vascular lesions, is low in fibrinolytic activity and high in thromboplastic activity, indicating an increased tendency to form and retain deposits of fibrin. The rat, on the other hand, has a coagulation inhibiting mechanism in its arteries. In this connexion it is of interest to recall that heparin and mucopolysaccharides with anticoagulant activity have been isolated from arteries of some species[14-16]. Apparently regulation of fibrin deposition can be obtained by several means depending upon the species in question. It is also of interest that high fibrinolytic activity (in man) is predominantly related to the venous system[17].

In none of the species investigated does the hæmostatic system compare exactly with that in the human organism. Based on our previous studies[9,10] the fibrinolytic system in human arteries appears similar to that in the pig, while the thromboplastic system is more active than in the rabbit. This means that the tendency to form fibrin deposits is more pronounced in man than in the rabbit, at the same time as there is little ability to produce resolution by means of a local fibrinolytic activity in the tissue. This finding suggests that resolution of fibrin deposits in the arterial system in man depends primarily on the activity of the humoral system in the circulating blood, which in man is characterized by a higher latent activity than in most animals. On the basis of these observations we offer the suggestion that the lack of a suitable experimental animal in studies of human arteriosclerotic lesions is due to differences in the hæmostatic systems of their arteries and in the circulating blood.

The results of this work will be published in detail elsewhere. Grants were received from the Josiah Macy, Jr., Foundation, New York, and from the National Danish Association against Rheumatic Diseases.

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[1] Duguid, J. B., *J. Path. Bact.*, **58**, 207 (1946); *Lancet*, ii, 925 (1949); i, 891 (1954); *Brit. Med. Bull.*, **11**, 36 (1955).
[2] Mallory, F. B., Harvey Lectures, 150 (1913).
[3] Clark, E., Graef, I., and Chasis, H., *Archiv Pathol.*, **22**, 183 (1936).
[4] Meyer, W. W., *Virchows Archiv.*, **314**, 681 (1947).
[5] Duguid, J. B., *J. Path. Bact.*, **60**, 57 (1948).
[6] Crawford, T., and Levine, L. I., *J. Path. Bact.*, **64**, 523 (1952).
[7] Astrup, T., *Lancet*, ii, 565 (1956); *Wiener Z. innere Medizin*, **39**, 373 (1958).
[8] Astrup, T., in Irvine Page, "Connective Tissue. Thrombosis and Atherosclerosis", 223 (Academic Press, New York, 1959).
[9] Astrup, T., and Claassen, M., Trans. Sixth Congress European Society Hæmatology, Copenhagen, 1957, 445 (Karger, Basle).
[10] Astrup, T., Albrechtsen, O. K., Claassen, M., and Rasmussen, J., *Circulation Res.*, **7**, 969 (1959).
[11] Witte, S., and Bressel, D., *Folia Hæmatologica*, **2**, 236 (1958).
[12] Campbell, L. L., *Amer. J. Pharmaceut. Assoc.*, **43**, 359 (1954).
[13] Astrup, T., and Albrechtsen, O. K., *Scand. J. Clin. Lab. Invest.*, **9**, 233 (1957).
[14] Jorpes, J. E., "Heparin", 56 (Oxford University Press, 1946).
[15] Berenson, G. S., *Biochim. Biophys. Acta*, **23**, 176 (1958).
[16] Kirk, J. E., *Nature*, **184**, 369 (1959).
[17] Todd, A. S., *Nature*, **181**, 495 (1958).

Effect of Cations on γ-Aminobutyric Acid Level in Slices of Brain Cortex

SINCE the discovery by Roberts and Frankel[1] of the unusually high content of γ-aminobutyric acid in brain, other mammalian tissues containing only minute amounts, a number of reports have appeared concerning the significance of this amino-acid to the physiological activity of the central nervous system[2].

The present communication deals with the effect of the concentration of potassium and calcium ions in the incubation media on the content of γ-aminobutyric acid in the slices. It is not without interest to examine the dependence of the amount of physiologically active γ-aminobutyric acid on the concentration of the ions K^+ and Ca^{2+} in the medium, as the importance of environmental ionic composition for the nervous tissue metabolism is known[3,4].

Guinea pigs were used as experimental animals; in individual experiments six groups of slices from four animals were prepared and the amount of the amino-acid in the tissue plus incubating medium after incubation in various salines was compared. Slices were incubated aerobically at 37° for 4 hr. in normal and modified Krebs–Ringer saline, the incubation stopped by denaturing the proteins, and the free amino-acids present in the supernatant after centrifugation of the homogenized slices separated chromatographically in a mixture of *n*-butanol/formic acid/water (75 : 15 : 10). Practically the same results were obtained with a 2-hr. incubation. The estimation of γ-aminobutyric acid in the corresponding strips of the chromatogram was carried out according to Yemm and Cocking[5].

It was found that, as compared with the control, only the intensity of the γ-aminobutyric acid spot on the chromatogram was markedly changed on

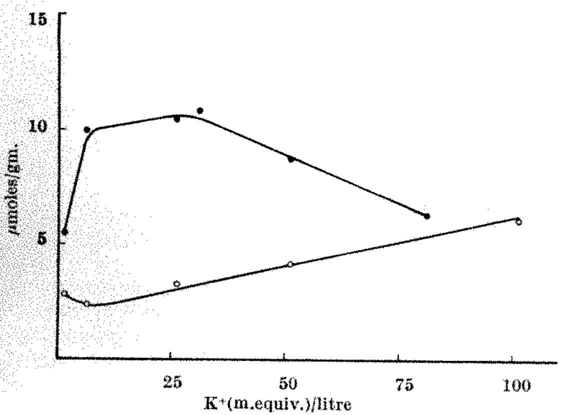

Fig. 1. Effect of K⁺ ions on free γ-aminobutyric acid and glutamine level in slices of brain cortex. The amounts of γ-aminobutyric acid (empty circles) and glutamine (full circles), estimated in the incubation media plus saline, expressed in μmoles/gm. fresh weight

incubation in a medium containing 25 m.equiv. K⁺/l. This observation was subjected to further analysis. As follows from Fig. 1, the γ-aminobutyric acid level is lowest during incubation in a medium containing 6·25 m.equiv. K⁺/l. With raised K⁺ concentration the amount of the amino-acid increases linearly and at 100 m.equiv. K⁺/l. it reaches the value of 6·2 μmoles/gm. fresh tissue. Also, the lowered potassium concentration causes an increase in the amino-acid level. The average amount of the amino-acid present in fresh slices was 2·9 μmoles/gm.

It is well known that γ-aminobutyric acid is formed by decarboxylation of glutamic acid. In another set of experiments under the same experimental conditions changes in the content of a different amino-acid, also arising metabolically from glutamic acid —namely glutamine—were followed. The amide was estimated from the amount of ammonia released on hydrolysis with trichloracetic acid, as described by Harris[6]. The decrease in glutamine formation, representing an endergonic reaction, with increasing amount of potassium in the incubation media is not surprising, the impairment of energy metabolism by potassium concentrations having been already repeatedly reported[3]. It appears (cf. Fig. 1) that no direct correlation exists between the γ-aminobutyric acid and glutamine levels in the slices.

Elliott and Florey[7] pointed out that a fraction of γ-aminobutyric acid is found in the brain tissue in a thermolabile 'bound' form hitherto not exactly defined. The question arose whether the increase in the γ-aminobutyric acid content was not partly due to the release of the acid from this labile form; therefore the amount of γ-aminobutyric acid was compared in boiled and unboiled homogenates after incubation in various media. It was found in preliminary experiments that the amount of this amino-acid was higher in the boiled homogenates after incubation of slices in the media with 1·25 and 6·25 m.equiv. K⁺/l. After incubation of brain tissue at higher concentrations of potassium the amount of the acid was nearly the same in boiled and in unboiled homogenates. It appears that the release of γ-aminobutyric acid from the thermolabile bond could be considered as a factor contributing to the increase in the level of the free amino-acid at higher concentrations of potassium in the saline. Furthermore, the possible activation of the glutamic acid decarboxylase under the experimental conditions described should also be taken into account.

On omitting calcium ions from the incubation medium the amount of γ-aminobutyric acid is increased, which is in agreement with the already well-established antagonistic action of calcium and potassium ions on brain cortex metabolism.

Details of this work will be published elsewhere.

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[1] Roberts, E., and Frankel, S., *J. Biol. Chem.*, **187**, 55 (1950).

[2] Purpura, D. P., Girado, M., and Grundfest, H., *Science*, **125**, 1200 (1957); *Proc. Soc. Exp. Biol. Med.*, **95**, 791 (1957). Iwama, K., and Jasper, H. H., *J. Physiol.*, **138**, 365 (1957).

[3] Ashford, C. A., and Dixon, K. A. C., *Biochem. J.*, **29**, 157 (1935). Findlay, M., Magee, W., and Rossiter, R. J., *ibid.*, **58**, 236 (1954). McIlwain, H., *ibid.*, **52**, 289 (1952). Kleinzeller, A., and Rybová, R., *J. Neurochem.*, **2**, 45 (1957).

[4] Vrba, R., Folbergrová, J., and Kanturek, V., *J. Neurochem.*, **2**, 187 (1958). Rybová, R., *ibid.*, **4**, 304 (1959).

[5] Yemm, E. W., and Cocking, E. C., *Analyst*, **80**, 209 (1955).

[6] Harris, M. M., *J. Clin. Invest.*, **22**, 569 (1943).

[7] Elliott, K. A. C., and Florey, E., *J. Neurochem.*, **1**, 181 (1956).

Attempted Extraction of Crustacean Moulting Hormone from Isolated Y-Organs

Echalier[1,2] has shown, by extirpation and implantation experiments, that the *Y*-organs of Crustacea[3] produce the moulting hormone. The *Y*-organs are therefore physiologically (and morphologically) homologous to the prothoracic glands of insects. It has also been demonstrated[4,5] that extracts from whole Crustaea (*Crangon vulgaris*[6], *Emerita talpodia*, *Libinia* spp.[7]) are active in insects, initiating puparium formation in ligated *Calliphora* larvæ. Thus, they have the same action as ecdysone, the moulting hormone or prothoracic gland hormone of insects.

We have been interested in learning whether the arthropod moulting hormones are stored in the glands of internal secretion, or if they are produced there and then released into the blood. Excision of prothoracic glands of insects would be extremely difficult; in contrast, the anatomy of the crustacean *Y*-organ allows preparation of the glandular tissue for chemical investigations.

100 *Y*-organs were dissected from green crabs (*Carcinus maenas*) of about 75-mm. carapace width, in stage C_4 (that is, the intermoult period) of the moulting cycle, and preserved in methyl alcohol. 36 carcasses were also stored. The glands were then homogenized in methanol, centrifuged and re-extracted with methanol. The methanolic extracts were combined, evaporated to a small volume, diluted with water and extracted first with ethyl ether, then with butanol; the butanol solution was taken to dryness, yielding 12·5 mgm. extract.

The carcasses were homogenized in a Waring blendor and extracted with methanol. The methanolic extracts (approx. 3 l.) were combined, concentrated to a small volume, and, after dilution with water, extracted first with ether, then with butanol; the butanol solutions were washed with 0·2 *N* sulphuric acid, 2 *N* sodium carbonate, water, 1 *N* acetic acid, and water, and evaporated to dryness. The residue (5·12 gm.) was dissolved in butanol, adsorbed on 50 gm. alumina (activity *V* according to Brockmann) and eluted with 200 ml. butanol (fract. 1), 200 ml.

Table 1

Starting material	Total final extract (mgm.)	Amount injected (μgm.)	Puparium formation (per cent)	*Calliphora* units (ref. 8)
100 *Y*-organs	12·5	250	0	<10
36 Carcasses without *Y*-organs	Fract. 1 127	50	58	2,500
	Fracts. 2–3 913	50	33	9,000

butanol (fract. 2) and 500 ml. butanol–methanol 1 : 1 (fract. 3). Fract. 1 (2·1 gm.) was dissolved in ether, then extracted with water, and the aqueous layer extracted with butanol. The yield, after evaporation of the solvent, was 127 mgm. Fracts. 2 and 3 were combined (1·15 gm.) and treated in the same way: yield, 913 mgm. The purified extracts were bioassayed in *Calliphora* larvæ (cf. ref. 8) (Table 1).

From the result of the bioassay, it is obvious that the *Y*-organs do not store measurable quantities of the hormone: 100 glands contain less than 10 *Calliphora* units, while 36 carcasses contain more than 10,000 units. These results were obtained using intermoult crabs; it remains to be seen whether the same relation holds for pre-moult crabs, and for insects. In the latter case, however, removal of prothoracic glands is not feasible because of their diffuse nature.

The positive response of *Calliphora* to *Carcinus* extracts demonstrates once more the interchangeability of insect and crustacean moulting hormones. There is little doubt that the *Calliphora* assay is reliable in this respect, though confirmation by assay in other insects, for example, *Rhodnius*[9] or *Cecropia*[10], might be desirable. The chemical properties of the crustacean hormone differ from the insect hormone: the former is more strongly adsorbed to alumina.

Another insect gland, the corpus allatum, which produces the juvenile hormone, contains undetectable amounts of active material. The juvenile hormone is either rapidly destroyed[11] or excreted[12] or piled up in the abdominal tissue[13]. Apparently, the endocrine glands of arthropods do not function as storage organs.

This work was supported by a grant from the Deutsche Forschungsgemeinschaft. Part of it was done during the tenure by one of us (P. K.) of a Lalor fellowship in the summer of 1956 and by the other (D. M. S.) of a U.S. Public Health Service predoctoral fellowship at Harvard University.

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[1] Echalier, G., *C.R. Acad. Sci., Paris*, **238**, 523 (1954).
[2] Echalier, G., *C.R. Acad. Sci., Paris*, **240**, 1581 (1955).
[3] Gabe, M., *C.R. Acad. Sci., Paris*, **237**, 1111 (1953).
[4] Karlson, P., *Ann. Sci. Nat. Zool. et Biol. Animale*, **18**, 123 (1956)
[5] Karlson, P., Proc. Fourth Internat. Congr. Biochem. 1958, **12**, 37 (Pergamon Press, London, 1959).
[6] Karlson, P., and Schmialek, P., *Hoppe-Seyler*, **316**, 183 (1959)
[7] Karlson, P., Report to Lalor Foundation 1957 (unpublished).
[8] Karlson, P., *Vitamins and Hormones*, **14**, 227 (1956).
[9] Wigglesworth, V. B., *J. Exp. Biol.*, **32**, 649 (1955)
[10] Williams, C. M., *Biol. Bull.*, **103**, 120 (1952).
[11] Gilbert, L. J., and Schneiderman, H. S., *Anat. Rec.*, **131**, 557 (1958).
[12] Karlson, P., and Schmialek, P., *Z. Naturforschung* (in the press).
[13] Williams, C. M., *Nature*, **178**, 212 (1956).

PLANT PHYSIOLOGY

Gravimorphism and Root Formation

IT has previously been reported[1,2] that gravity can affect the growth and flowering of trees. The results to be described indicate that gravity may also affect the growth of some herbaceous plants in that the orientation of the shoot can determine whether or not root initials are formed in the stem.

Pot-grown plants of *Bryophyllum daigremontianum* (R. Hamet et Perv.) Berg. and tomato var. 'Moneymaker' were placed so that the shoots and the pots were horizontal. Staking ensured that the shoots remained horizontal within practical limits. One group of horizontal plants had the same side uppermost throughout, while a second group was placed on a horizontal klinostat which rotated the plants approximately once an hour. Preliminary experiments showed that rotating the plants through 180° once or twice each day did not prevent the formation of adventitious roots. A third group of plants was maintained with the stems and pots vertical. To facilitate watering, all the horizontal plants were placed erect for a short period each day.

After a few weeks of such treatments, adventitious roots were clearly visible on the underside of the stem internodes of those plants that were maintained with the same side uppermost. In *B. daigremontianum* these adventitious roots were visible after 2–3 weeks and continued to increase in size and number during the ensuing weeks (Fig. 1). At the end of the experiment many of the roots exceeded 2·0 cm. in length. Root formation in tomato was slower, taking 4–5 weeks to become visible. Under the conditions of the experiment tomato roots did not extend, although the lower side of the stems became covered with numerous root primordia. In both species roots were confined to the lower side of the horizontal stems. The rotated and vertical plants did not exhibit any abnormal propensity to form roots. Table 1 summarizes the results of one experiment with *B. daigremontianum*. The undersides of the horizontally placed stems were shaded, and it is not known how much this differential illumination may have affected the results[3,4]. This will be investigated and reported on in due course.

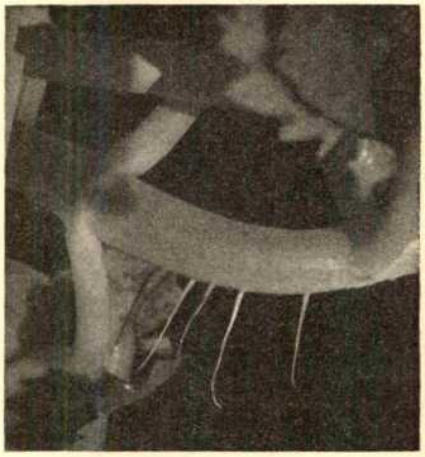

Fig. 1. Adventitious roots arising from a horizontally placed stem of *Bryophyllum daigremontianum*

Table 1. THE EFFECT OF GRAVITY ON THE ADVENTITIOUS ROOT FORMATION OF *Bryophyllum daigremontianum*

Stem position	Duration of experiment	No. of plants	No. of visible adventitious roots
Horizontal, rotated	37 days	6	2
Horizontal, non-rotated		6	72
Vertical		6	0

Loeb[5] observed that ". . . in *Bryophyllum calycinum* the substances which induce root formation have a tendency to collect on the lower side of a horizontally placed stem although roots may also appear in the nodes on the upper side. . . ." All the roots in Loeb's drawing were of nodal origin and not predominately internodal as in the present experiments. Resende[6], working with *B. daigremontianum*, noted that roots formed on the stems of horizontally placed plants at the point where the stems curved as a result of geotropic action. Both authors appear to have been recording manifestations of gravimorphism similar to those already described.

These observations are of horticultural significance in relation to the practice of layering used for the propagation of difficult subjects. Further, the rapid response of *B. daigremontianum* presents a ready means of demonstrating one of the effects of gravimorphism to students.

A more detailed account of this work will be published elsewhere.

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[1] Wareing, P. F., and Nasr, T., *Nature*, **182**, 379 (1958).
[2] Longman, K. A., and Wareing, P. F., *Nature*, **182**, 380 (1958).
[3] Toyomura, S., and Yosioka, S., *Bul. sci. Fak. Terkult. Kyusu imp. Univ.*, **8**, 204 (1939).
[4] Viana, M. J., *Portugal. Acta biol.*, Ser. A, **3**, 195 (1951).
[5] Loeb, J., *Bot. Gaz.*, **63**, 25 (1917).
[6] Resende, F., *Portugal. Acta biol.*, Ser. A, **2**, 251 (1948).

Fungitoxicity of Metal Ions

IN a recent communication, Somers[1] has again considered the hypothesis first proposed by Mathews[2], that the toxicity of metal ions is related to the electrode potential of the metal. He has plotted the negative logarithms of the *ED*50 values of the applied molar concentrations of 21 elements for conidia of *Alternaria tenuis* against the electronegativity of the elements. An examination of the curve indicates clearly a correlation between the electronegativity and toxicity in the data presented. This is especially fortified by the low electronegativity of magnesium, strontium, lithium, sodium, barium and potassium ions which are basically not toxic to fungus conidia. If, however, attention is confined to that part of the curve which deals with ions toxic to fungus conidia (ions as toxic and more toxic than zinc), it is apparent that in a number of instances, the *ED*50 values are several orders of magnitude higher or lower than if they fell on the line drawn. As a matter of fact, examination of the data for these 15 elements, by calculating the rank correlation coefficient (calculated by Dr. F. Wilcoxon, Boyce Thompson Institute), shows that there is no significant correlation between electronegativity and toxicity in the area in which the toxic elements are found.

It should be pointed out that much information has been accumulated in recent years showing that the concentration of the applied solution is a poor measure of toxicity[3-9]. Fungus conidia have a great affinity for toxic ions and compounds as a result of which they often will take up lethal doses from dilute solutions in a minute or less. The applied concentration is thus maintained for a short time only and is important, not as such, but because it determines the dose available per unit of fungus conidia. Two toxicants may give equal *LD*50 values on an applied concentration basis but yet differ a hundredfold or more in innate toxicity. This follows because the germination response is determined by the quantity of toxicant taken up by the conidia with little regard to how such uptake was brought about. This is true for most of the toxicants studied, although there are exceptions. With zinc and cadmium the toxic effects are influenced by the length of exposure to the toxicant solution even when similar amounts are taken up[7].

Obviously the amount of a given ion taken up by conidia depends upon the quantities of binding sites in the spores and their availability to the ions presented in the ambient solution. The resultant toxicity would be determined by the essential nature of the binding sites occupied, and reversibility would be possible if the bond between the toxicant and the binding sites can be broken without impairment of essential functions. It is apparent, as also brought out by Somers[1], that chemical reactions occur between the applied ions and cell constituents. Such reactions can occur after the conidia are no longer capable of germination and thus can account for uptake of toxicants by spores no longer viable[4].

The question arises as to where these reactions between binding sites and the applied ions occur. Somers[1], in common with some others, believes that they occur at the cell surface. This point of view results from a belief that the conidia are permeable with difficulty to ions and compounds supplied and, moreover, it assumes that enough of the essential processes of the cell takes place at the surface, so that reaction at the surface can destroy germinative capacity. Experiments conducted at the Boyce Thompson Institute over the past ten years with labelled ions and compounds and labelled cell constituents have shown that a much freer movement of materials takes place across the membranes involved than was previously supposed. If fungus spores are collected without immersion in water and are then suspended in water, from 10 to more than 30 per cent of the cell constituents is released into the ambient solution[10]. The remaining contents are more firmly held, but further release will take place, without impairment of germination, on exposure to low doses of certain potentially toxic agents such as silver. Silver has been found to affect the conidia in such a way as to bring about a release of phosphorus compounds, as determined with spores labelled with phosphorus-32 [6,7]. Effects of silver on permeability are also shown by its action in increasing the uptake of other ions supplied, such as mercury and cerium[5].

Sonic disintegration of spores previously treated with relatively low doses of silver, mercury, cadmium, zinc, or cerium, and determination of the distribution of the labelled ions in various fractions isolated have not indicated an accumulation of these elements at the cell surface[11]. While there is a possibility of some redistribution taking place during the disintegration process, experiments indicate that very little redistribution actually occurs.

When conidia of different species are compared the quantity of toxicant that can be taken up is found to vary greatly from species to species. A species taking up very large amounts of one toxicant will not necessarily take up comparatively large amounts of a second toxicant. Also the ion may not be very toxic to a species which has a great affinity for it.

Experiments being carried out with cerium have shown that with some species the binding sites for cerium are readily available to cerium supplied in the ambient solution. With others the addition of 'Tergitol-7' (sodium heptadecyl sulphate) or CN^-, in non-toxic quantities, makes available further binding sites and thereby increases cerium uptake. The total potential cerium uptake, of course, also varies greatly among the species studied.

Ion interference studies now being conducted indicate that the interaction between conidia and the metal ions is much more complex than can be explained by differences in electronegativity. Thus cerium would not be expected to compete with silver or mercury for the same receptor sites, yet small amounts of cerium seriously interfere with the uptake of silver and mercury.

The evidence strongly indicates that much more than cell surface reactions is involved in the toxicity of metal ions to conidia.

Unpublished results referred to above will be reported in detail elsewhere. These investigations were supported in part by the United States Atomic Energy Commission and the Rockefeller Foundation.

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[1] Somers, E., *Nature*, **184**, 475 (1959).
[2] Mathews, A. P., *Amer. J. Physiol.*, **10**, 290 (1904).
[3] Miller, L. P., McCallan, S. E. A., and Weed, R. M., *Contrib. Boyce Thompson Inst.*, **17**, 173 (1953).
[4] Miller, L. P., McCallan, S. E. A., and Weed, R. M., *Contrib. Boyce Thompson Inst.*, **17**, 283 (1953).
[5] Miller, L. P., McCallan, S. E. A., and Weed, R. M., Proc. Second Radioisotope Conf., Oxford, **1**, 381 (1954).
[6] Miller, L. P., McCallan, S. E. A., Proc. Intern. Conf. on Peaceful Uses of Atomic Energy, **12**, 170 (1956).
[7] Miller, L. P., and McCallan, S. E. A., *J. Agric. Food Chem.*, **5**, 116 (1957).
[8] Burchfield, H. P., and Storrs, E. E., *Contrib. Boyce Thompson Inst.*, **18**, 429 (1957).
[9] Owens, R. G., and Novotny, H. M., *Contrib. Boyce Thompson Inst.*, **20**, 151 (1959).
[10] Miller, L. P., and McCallan, S. E. A., *Science*, **126**, 1233 (1957).
[11] Owens, R. G., and Miller, L. P., *Contrib. Boyce Thompson Inst.*, **19**, 177 (1957).

Hydrogenation of Chloroplast Lipids by Rumen Bacteria

It was reported in an earlier paper[1] that rumen protozoa can hydrogenate unsaturated lipids. The suggestion was made that rumen bacteria may not contribute a great deal to hydrogenation of dietary lipids ingested by ruminants. This latter conclusion was based on experiments using washed suspensions of rumen bacteria, incubated anaerobically in phosphate buffer containing sodium sulphide, with either linseed oil, or linoleic or linolenic acids. Formic acid, glucose or finely divided plant material were also added in these experiments. Estimation of the iodine numbers of the total fatty acids in the samples showed either no change or only a slight decrease in the iodine values.

Since these results were considered rather unexpected, the role of the bacteria in hydrogenation has been reappraised using chloroplasts as a source of unsaturated lipid substrate. Rumen liquor was obtained from a rumen-fistulated cow routinely fed on fresh red clover (*Trifolium pratense*), care being taken to maintain both the temperature of the sample and anaerobic conditions. Washed suspensions of the bacteria were prepared by a method similar to that described by Doetsch, Shaw, McNeill and Jurtshuk[2]. The suspension was divided into two equal portions, one of which was placed in hot water (90°) for 30 min. to destroy enzyme activity. Equal aliquots of a chloroplast suspension[1] together with glucose (0·25 gm./100 ml.) were added to both samples and the flasks incubated overnight at 39° under carbon dioxide.

A second similar experiment was carried out using strained rumen liquor from which the protozoa and plant material had been removed by centrifugation (1,000*g* for 5 min.). Chloroplasts and glucose were added to the suspensions, one of which had been heated as above, and the flasks incubated overnight at 39° under carbon dioxide.

The samples were freeze-dried and the lipids extracted by boiling successively with acetone, absolute ethanol and diethyl ether. The solutions were evaporated to dryness, taken up in petroleum ether and the solvent removed *in vacuo*. The fatty acids from the lipids were isolated by the usual procedures[1] and their methyl esters analysed by gas-liquid chromatography.

Table 1. CARBON-18 FATTY ACID COMPOSITION OF CHLOROPLAST LIPIDS INCUBATED WITH RUMEN BACTERIA

Sample + chloroplasts + glucose (0·25 per cent w/v)	Suspending medium	Fatty acids (moles per cent)			
		Saturated	Unsaturated		
			mono-	di-	tri-ene
Live bacteria	*M*/15 phosphate buffer (*p*H 7·0) + 0·02 per cent $Na_2S.9H_2O$	4·1	11·0	7·6	77·3
Heated bacteria		4·0	4·0	10·0	82·0
	Difference	+0·1	+7·0	−2·4	−4·7
Live bacteria	Rumen liquor *p*H 7·0	9·4	24·1	62·2	4·3
Heated bacteria		5·0	6·5	5·0	83·5
	Difference	+4·4	+17·6	+57·2	−79·2

The results (Table 1) show a great contrast in hydrogenation activity between the bacteria suspended in phosphate buffer and those in rumen liquor. The bacteria incubated in phosphate buffer have caused only a slight, although definite, hydrogenation of the unsaturated fatty acids contained in the chloroplasts. There has been some conversion of triene to diene and monoene acids in the live bacteria sample. The bacteria suspended in rumen liquor have converted nearly all the triene acid to more saturated acids, particularly the diene acids. It is interesting that there is a build-up of the diene acids as this corresponds to the results found with rumen protozoa when it was suggested that the enzymes responsible for hydrogenation showed some specificity and attacked the triene acids more than the diene acids.

The low degree of hydrogenation in the washed suspensions is presumably the result of either harmful effects caused in the handling and preparing of these suspensions or the presence of some factor in rumen fluid that facilitates hydrogenation and explains the previous conclusions regarding the role of bacteria in ruminant hydrogenation. However, these results show there can be no doubt that rumen bacteria are capable of extensive hydrogenation, and emphasize the caution needed in the interpretation of results obtained under conditions far removed from those found *in vivo*.

I wish to thank Miss J. Michael for her able assistance and the Fats Research Laboratory, Wellington, for the gas–liquid chromatography analyses.

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[1] Wright, D. E., *Nature* (in the press).

[2] Doetsch, R. N., Shaw, J. C., McNeill, J. J., and Jurtshuk, P., University of Maryland, Misc. Pub., 238 (1955).

HÆMATOLOGY

Red Cell Agglutination by Stromal Material in Hæmolysates

During the preparation of hæmoglobin solutions a phenomenon was observed which, so far as is known, has not previously been described. The red cells of some species of animals are agglutinated by hæmolysates made from the cells of the same species. The degree of agglutination is sufficiently strong to be easily visible to the naked eye when tested on a slide.

Hæmolysates were prepared from the cells of a number of different animals by subjecting a 20 per cent suspension of washed cells to ultrasonic vibration. After centrifuging for 20 min. at 3,000 rev./min. to remove the larger particles of stroma, it was tested against washed cells of the various species. The results are shown in Table 1, where the degree of agglutination is recorded, following the usual practice of blood transfusion work[1]. From Table 1 it can be seen that the agglutination seems to be a property of the hæmolysate rather than of the intact cell; and that it is confined to the three rodent species.

Table 1

Cells of	Hæmolysates of cells of: Sheep	Rabbit	Dog	Horse	Rat	Guinea pig
Sheep	0	+	0	0	0	+
Rabbit	0	+++	0	0	++++	++
Dog	0	0	0	0	+	++
Horse	0	++++	0	0	0	0
Rat	0	+	0	0	+++	+
Guinea pig	0	0	0	0	+	+

Group *A* and *O* human cells were not agglutinated by any hæmolysate and hæmolysates of human cells of groups *A* and *O* caused no agglutination of cells of any species.

If packed washed rabbit cells are frozen and thawed with the minimum of disturbance a jelly-like material with the consistency of egg-white is produced. This material, which is probably stromatin[2], can be filtered off and washed. The filtered hæmoglobin, although very concentrated, produces no agglutination of intact red cells. On the other hand, the washed residue, although not entirely free of hæmoglobin, is very active in producing agglutination when dissolved in saline. This property can be removed by absorption with rabbit cells but is unaffected by treatment with fat solvents.

The properties of the hæmolysates are dependent on the method of preparation. Fresh washed rabbit cells were lysed: (1) by ultrasonic vibration; (2) by the addition of distilled water followed by the calculated quantity of 10 per cent saline to make the solution isotonic and (3) by freezing. The hæmolysates were handled with care to avoid shaking. On completion of lysis all three preparations produced a similar degree of agglutination. After centrifuging at 96,000*g* for 1 hr. it was found that the supernatant fluids varied considerably in their ability to agglutinate washed cells. The deposit was re-suspended in saline by shaking vigorously for at least 5 min. before centrifuging at 3,000 rev./min. for ½ hr. The results of testing the supernatant fluid and the deposit against whole cells are summarized in Table 2.

Table 2

Method of production of hæmolysate	Hæmolysate: Deposit	Supernatant
1. Ultrasonic vibration	+	+++
2. Distilled water	+++	+
3. Freezing	+++	0

The probable explanation of these findings is that the more violent method of disruption of the cells also disintegrates the red cell envelope and stroma, some product of which is added to the solution and not removed by high-speed centrifugation for 1 hr. The gentler methods, however, leave this substance attached to the 'cell ghosts' which are found in the deposit.

We can conclude, therefore, that the agglutination of homologous and some heterologous red cells by hæmolysates of rabbit, rat and guinea pig red cells is due to a substance liberated from the stroma. The amount of this substance in the solution is determined by the method of lysis and the amount of shaking during preparation. This variation in composition may explain the divergent results obtained by the earlier workers in experimental hæmoglobinuric nephrosis.

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[1] Stratton, F., and Renton, P. H., "Practical Blood Grouping" (Blackwell, Oxford, 1958).

[2] Ponder, E., "Hemolysis and Related Phenomena" (Grune and Stratton, New York, 1944).

Asymmetrical Recombination of Alkali-dissociated Hæmoglobin Mixtures

The human adult hæmoglobins and canine hæmoglobin recombine asymmetrically, that is:

$$\alpha_2^X\beta_2^X + \alpha_2^Y\beta_2^Y = \alpha_2^X\beta_2^Y + a_2^Y\beta_2^X$$

after both short and prolonged exposure to dilute acid[1-5]. At *p*H 11, hæmoglobin *A* dissociates into half-molecules[6]; furthermore, re-association of a mixture of labelled hæmoglobin *S* and hæmoglobin *A* after 24 hr. at *p*H 11 results in the exchange of the β_2 sub-units[7]. We now wish to report a series of experiments which indicate that hæmoglobin also recombines asymmetrically after short exposure to alkali.

In the first experiment a 2 per cent carbonmonoxyhæmoglobin solution (all carbonmonoxyhæmoglobin

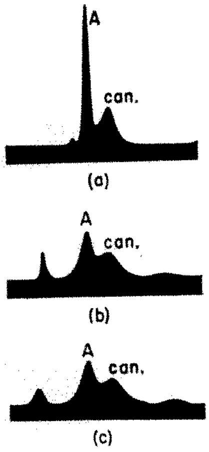

Fig. 1

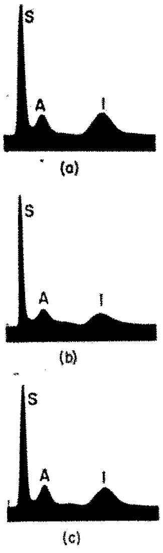

Fig. 2

Fig. 1. Ascending-limb electrophoretic patterns of 1 per cent carbomonoxyhæmoglobin in phosphate buffer of pH 6·8, Γ/2 0·02. The direction of migration is from right to left. (*a*) *A*, canine control; (*b*) *A*, canine recombined after 20 min. dissociation at pH 11·2; (*c*) *A*, canine recombined after 20 min. dissociation at pH 4·7

Fig. 2. Ascending-limb electrophoretic patterns of 1 per cent. carbomonoxyhæmoglobin in phosphate buffer of pH 6·3, Γ/2 0·02. The direction of migration is from right to left. (*a*) *S,A,I*, control; (*b*) *S,A,I*, recombined after 20 min. dissociation at pH 11·2; (*c*) *S,A,I*, recombined after 20 min. dissociation at pH 4·7. Note the new component between *A* and *I* in (*b*) and (*c*)

solutions were electrodialysed prior to use), 1 per cent in canine hæmoglobin and 1 per cent in human hæmoglobin, was diluted to 1 per cent by adding an equal volume of a glycine–sodium hydroxide buffer of pH 11·0, Γ/2 0·2. The solution was dialysed against this same buffer at Γ/2 0·1 for 24 hr. at 5° C. followed by dialysis against three changes of potassium phosphate buffer of pH 6·8, Γ/2 0·02 for approximately 17 hr. Upon electrophoresis in the same phosphate buffer two new components appeared, one migrating more slowly than canine and the other migrating more rapidly than *A*, as was the case after exposure to dilute acid (Fig. 1). 20-min. exposure to the same glycine–sodium hydroxide buffer produced only very small amounts of the new components. It was found, however, that the addition of the hæmoglobin mixture to the glycine–sodium hydroxide buffer changed the pH to 10·5 and therefore further experiments were carried out with glycine–sodium hydroxide buffers of a higher pH. A buffer of pH 11·5, Γ/2 0·2, when added to an equal volume of 2 per cent carbonmonoxyhæmoglobin, resulted in a pH of 11·0, and a buffer of 11·8 resulted in a final mixture pH of 11·2. After 20-min. exposure, the amounts of the new components observed upon electrophoresis at pH 6·8 were found to increase with increase in the dissociation pH. In one experiment in which the pH 11·8 buffer was used, immediate neutralization to pH 6·9 was effected after 20 min. by the addition of sodium dihydrogen phosphate. In all the others, 20-min. exposure to alkali was followed by dialysis in the potassium phosphate buffer pH 6·8. These results are shown in Table 1 and Fig. 1. Human and canine carbonmonoxyhæmoglobin, when dissociated separately at pH 11·0 for 20 min. and re-associated, showed no change in electrophoretic composition. Dissociation and recombination did not alter the absorption spectrum (450–700 mμ) of any of the samples.

Table 1. COMPOSITION OF CANINE AND HUMAN HÆMOGLOBIN MIXTURES BEFORE AND AFTER RECOMBINATION

Dissociation (pH)	Dissociation (time)	Electrophoretic composition (per cent of total)*			
		New slow	Canine	*A*	New fast
Control	—	—	37	63†	—
10·5	20 min.	2–3	39	55	4
11·0	20 min.	5	37	51	7
11·2	20 min.	11	35	41	13
11·2	20 min.‡	12	36	42	10
11·0	24 hr.	25	26	20	29
4·7	20 min.	10	36	44	10
4·7	22 hr.	31	24	21	24

* All samples contained 50 per cent each of human and canine hæmoglobin before recombination.

† Includes 2 per cent A_2.

‡ Followed by rapid neutralization to pH 6·9 by addition of sodium dihydrogen phosphate.

A 50–50 mixture of carbonmonoxyhæmoglobins *A* and *I* (the sample contained 40 per cent *A* and 60 per cent *I*) and carbonmonoxyhæmoglobin *S* was dissociated at pH 11·2 for 20 min., neutralized rapidly to pH 6·9 by the addition of sodium dihydrogen phosphate and dialysed against three changes of potassium phosphate buffer of pH 6·3, Γ/2 0·02 for about 18 hr. Moving-boundary electrophoresis in the same buffer showed a new component migrating between *A* and *I*, a decrease in the proportions of *S* and *I* and increase in the proportion of *A*. The dissociation and recombination of *A* and *I* alone produced no change in the electrophoretic pattern. The results are given in Table 2 and Fig. 2.

Table 2. COMPOSITION OF *A*, *I* AND *S* HÆMOGLOBIN MIXTURES BEFORE AND AFTER RECOMBINATION

Mixture	Dissociation (pH)	Dissociation (time)	Electrophoretic composition (per cent of total)			
			I	New	*A*	*S*
$A + I$	Control	—	54	—	46	—
$A + I$	11·2	20 min.	56	—	44	—
$A + I + S$*	Control	—	30	—	17†	53
$A + I + S$	11·2	20 min.‡	25	7	18	50
$A + I + S$	4·7	20 min.§	29	4	18	49

* All $A + I + S$ samples contained 50 per cent hæmoglobin *S* and 50 per cent $A - I$ hæmoglobin mixture, the latter containing 40 per cent *A* and 60 per cent *I*.

† Includes 1 per cent of trailing shoulder.

‡ Followed by rapid neutralization to pH 6·9 by addition of sodium dihydrogen phosphate.

§ Followed by rapid neutralization to pH 6·4 by addition of disodium hydrogen phosphate.

An attempt to perform similar experiments with human hæmoglobin *A* using the 'hæm-labelling method'[2] was unsuccessful, due to the rapid denaturation of methæmoglobin at pH 11 and above.

The appearance of two new electrophoretic components after alkalinization and neutralization of a mixture of human hæmoglobin *A* and canine hæmoglobin parallels the findings after exposure to dilute acid. The net change can be represented by the equation:

$$\alpha_2{}^{A}\beta_2{}^{A} + \alpha_2{}^{\mathrm{can}}\beta_2{}^{\mathrm{can}} = \alpha_2{}^{\mathrm{can}}\beta_2{}^{A} + \alpha_2{}^{A}\beta_2{}^{\mathrm{can}}$$

Similarly, the appearance of a new component between *A* and *I* and an increase in the proportion of *A* after alkali dissociation and neutral re-association of a mixture of hæmoglobins *A* and *I* and hæmoglobin *S* parallels the findings after acidification. The net change in this case can be represented by the equation:

$$[\alpha_2{}^{I}\beta_2{}^{A}]^{-4} + [\alpha_2{}^{A}\beta_2{}^{S}]^{+2} = [\alpha_2{}^{I}\beta_2{}^{S}]^{-2} + [\alpha_2{}^{A}\beta_2{}^{A}]^{0}$$

It would appear that the mode of recombination of a mixture of hæmoglobins after exposure to dilute alkali is the same as that following exposure to dilute acid, that is, asymmetric.

Although at equilibrium carbonmonoxyhæmoglobin *A* is completely dissociated at *p*H 11 and but partially dissociated at *p*H 4·7 (ref. 6), our results show that the amount of asymmetric recombination is the same after 20 min. at either *p*H. Complete recombination occurs in either case after 22–24 hr. The extent to which partial recombination occurs after a short period of dissociation thus appears to reflect more closely the rate of exchange of sub-units rather than the equilibrium degree of dissociation.

Acidification of hæmoglobin results in asymmetric dissociation into α_2 and β_2 sub-units[2]. Asymmetric dissociation in alkali would provide the simplest mechanism compatible with our present results. If, as proposed by Vinograd and Hutchinson[8], hæmoglobin dissociates symmetrically into the half-molecule $\alpha\beta$ in alkali, the mechanism of recombination in alkali must be more complex than in acid.

We are indebted to Dr. L. M. Tocantins for the sample of *A–I* hæmoglobin.

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[1] Itano, H. A., and Singer, S. J., *Proc. U.S. Nat. Acad. Sci.*, **44**, 522 (1958).

[2] Singer, S. J., and Itano, H. A., *Proc. U.S. Nat. Acad. Sci.*, **45**, 174 (1958).

[3] Itano, H. A., Singer, S. J., and Robinson, E., Ciba Found. Symp. Human Biochemical Genetics (Churchill, London, 1959; in the press).

[4] Itano, H. A., and Robinson, E., *Nature*, **183**, 1799 (1959).

[5] Itano, H. A., and Robinson, E., *Nature*, **184**, 1468 (1959).

[6] Hasserodt, U., and Vinograd, J., *Proc. U.S. Nat. Acad. Sci.*, **45**, 12 (1959).

[7] Vinograd, J. R., Hutchinson, W. D., and Schroeder, W. R., *J. Amer. Chem. Soc.*, **81**, 3168 (1959).

[8] Vinograd, J. R., and Hutchinson, W. D., unpublished work cited by Kendrew, J. C., *Fed. Proc.*, **18**, 740 (1959).

PATHOLOGY

Plasma Erythropoietin in Polycythæmia Secondary to Renal Disease

An increasing body of experimental work indicates an intimate relationship between the kidney and the plasma erythropoietin-level[1-4]. It has long been recognized that in man there is a frequent association between chronic renal disease and anæmia, and it has been demonstrated that the plasma erythropoietin-level is reduced in chronic renal failure[5]. More recently, the association of certain other renal pathological conditions (for example, carcinoma of the kidney and hydronephrosis) with polycythæmia has been recognized[6,7]. However, studies on the plasma erythropoietin-level do not appear to have been reported in polycythæmia secondary to renal disease.

This communication is a preliminary report of studies on two such cases (to be reported more fully at a later date).

The two patients were middle-aged men in whom the diagnosis of polycythæmia was confirmed by red cell volume studies using chromium-51. The renal lesions were unilateral hydronephorosis in case I and a unilateral polycystic kidney in case II. In both cases, nephrectomy was performed, and the studies were repeated approximately four months after operation, at which time they were no longer polycythæmic.

Table 1

		Hb (gm. per cent)	P.C.V. (per cent)	Reticulocytosis in rats 72 hr. after injection of plasma (per cent)
Case I	Preoperative	18·3	55·0	5·7 6·1 Mean = 5·9
	16 weeks after operation	14·6	44·5	3·4 3·3 Mean = 3·4
Case II	Preoperative	18·7	55·0	8·9 8·4 Mean = 8·7
	19 weeks after operation	15·2	47·5	3·0 3·3 Mean = 3·2
Pooled plasma from five normal subjects		—	—	3·3 3·2 Mean = 3·3

Plasma samples were obtained pre-operatively and approximately four months post-operatively. The erythropoietin content of these samples was assessed by measuring the reticulocytosis induced, in black hooded rats, 72 hr. after a 2 ml. intraperitoneal injection of plasma. The results are given in Table 1.

These two cases appear to demonstrate convincingly the association of certain renal lesions with polycythæmia and increased plasma erythropoietin-levels, and show that a return to the normal erythropoietic state follows removal of the pathological kidney.

We wish to express our thanks to Dr. J. L. Pinniger for helpful advice and for permission to publish this communication.

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[1] Jacobson, L. O., Goldwasser, E., Fried, W., and Plzak, L., *Nature*, **177**, 1240 (1957).

[2] Naets, J. P., *Nature*, **181**, 1134 (1958).

[3] Osnes, S., *Brit. Med. J.*, ii, 1387 (1958).

[4] Lange, R. D., Gallagher, N. I., McCarthy, J. M., and Wright, S., *J. Lab. Clin. Med.*, **53**, 912 (1959).

[5] Gallagher, N. I., McCarthy, J. M., Hart, K. T., and Lange, R. D., *Blood*, **14**, 662 (1959).

[6] Damon, A., Holub, D. A., Melicow, M. M., and Uson, A. C., *Amer. J. Med.*, **25**, 182 (1958).

[7] Forssell, J., *Acta Med. Scand.*, **161**, 169 (1958).

Identification of a Bovine Mucosal-Disease Virus isolated in Sweden as *Myxovirus parainfluenzae 3*

'Bovine mucosal disease' is the collective name proposed for different syndromes characterized mainly by inflammatory changes in the mucosa of the gastro-intestinal or respiratory tracts, or both, of cattle. The ætiology was uncertain in spite of successful transmission experiments. By the use of tissue culture for virus isolation, what are apparently different viruses have been found associated with such syndromes, but only one of these, the virus of infectious bovine rhinotracheitis, has hitherto been investigated thoroughly.

Quite recently, Reisinger *et al.*[1] in the United States reported the isolation of a myxovirus from cases of shipping fever, a respiratory disease in cattle. Abinanti and Huebner[2] found this virus, called *SF*–4, to be serologically identical to the prototype strain of *Myxovirus para-influenzae* 3 (ref. 3), called *HA*–1, which was first isolated by Chanock *et al.*[4] from cases of respiratory disease in children.

A contagious form of enteritis in cattle, which seemed to be a variant of the mucosal disease type, has been described from Sweden[5]. The clinical signs have varied from outbreak to outbreak; the gastro-intestinal signs may dominate in one outbreak and respiratory signs in another. Differences in clinical signs were also observed within herds and during an outbreak[6].

Bakos *et al.*[7] investigated cases of a bovine mucosal-type disease with mainly respiratory symptoms and a severe course at Umeå in northern Sweden during 1958–59. The disease seemed to be introduced by cattle purchased from other parts of the country. The first signs of the disease were serous conjunctivitis and rhinitis, often combined with excessive salivation, high fever and, in some animals, diarrhœa. Then the nasal discharge became muco-purulent, the salivation more intensive and buccal erosions sometimes appeared; coughing was common. In some instances watery diarrhœa was the only sign encountered. The malignant course led in one of the infected herds of losses of up to 70 per cent of animals by inanition and dehydration. A cytopathogenic agent was isolated in calf kidney tissue cultures from nasal and tracheal exudates of the animals in two of the nine herds investigated. This agent was neutralized by convalescent sera from animals in these herds. It passed 'Gradocol' membranes with an average pore diameter of 240 mμ but not 130 mμ. In calves free of neutralizing serum antibodies, the agent produced in experimental infection a mild form of mucosal disease of the same type seen on the farms.

At titration of this so-called Umeå-strain of virus in calf kidney tissue cultures, it showed only slight cytopathogenicity but was more cytopathogenic when inoculated at higher dilutions. Therefore the subsequent passages were carried out in two lines, one with undiluted inocula and the other diluted 10^{-2} before inoculation. The former has been carried for 23 passages and the latter for 33 passages. The viruses have been designated strain 23 and 33 respectively. Both strains showed hæmagglutinins against guinea pig and chicken erythrocytes and strong hæmadsorption of guinea pig erythrocytes to the tissues in the infected tubes. Of the strains 23 and 33, only the latter produced clear plaques.

The strains 23, 33 and Chanock's[4] strain *HA*–1 of para-influenza 3 virus[3] have been compared by titration in calf and monkey kidney tissue cultures using lactalbumin-Hanks' solution (0·5/99·5 per cent) as maintenance medium. The hæmadsorption and hæmadsorption-inhibition tests were performed according to Chanock's modification[4] of the method described by Shelokov *et al.*[8], and the hæmagglutination, hæmagglutination-inhibition, and neutralization tests according to the standard techniques. In *HI* tests, the Umeå serum has been used untreated, and treated with *RDE* (cholera filtrate, Philips-Roxane, Holland). No difference could be observed in the antibody titres between the treated and untreated samples. In some of the experiments the *HA*-2 strain[4] of para-influenza 1 virus[3] and a serum against this strain were used for comparison.

The strain 23 showed slight or questionable cytopathogenicity for calf kidney cells only, and lower titres of infectivity for calf and monkey kidney cells than the strains 33 and *HA*–1. The strain 33 was definitely cytopathogenic for calf kidney cells and the strain *HA*–1 for monkey kidney cells, both showing approximately the same titres of infectivity. In neutralization tests monkey kidney cells were used for the strain *HA* 1 and calf kidney cells for the strain 33.

The results of cross-neutralization tests, summarized in Table 1, indicate a close relationship between the Umeå virus and the *HA* 1 strain of para-influenza 3 virus.

Table 1. EVIDENCE FOR THE SEROLOGICAL RELATIONSHIP BETWEEN THE VIRIS UMEÅ AND THE STRAIN *HA* 1 OF PARA-INFLUENZA 3 VIRUS

Serum	Hæmadsorption inhibition[a]				Hæmagglutination inhibition[b]			Neutralization[c]	
	23	33	*HA* 1	*HA* 2	23	33	*HA* 1	33	*HA* 1
Umeå[d]	+	+	+	–	4096[e]	2048	1024	128[e]	512
HA 1	+	+	+	–	1024	1024	1024	256	1024
HA 2	–	–	–	+	<16	<16	<16	ND	ND

[a] Infected monkey or calf kidney tissue cultures ;
[b] Against 4 hemagglutinating units of each virus ;
[c] Against 1,000 and 700 TCD_{50} of 33 and *HA* 1, respectively ;
[d] Convalescent serum from a calf (No. 1) experimentally infected with the virus Umeå and with no antibodies before infection ;
[e] Reciprocal of serum dilution ;
ND, not done.

We are indebted to Dr. L. Philipson, of the Institute of Virology, Uppsala, for supplying the reference viruses and sera *HA* 1 and *HA* 2.

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---

[1] Reisinger, R. C., Heddleston, K. L., and Manthei, C. A., *J. Amer. Vet. Med. Assoc.*, **135**, 147 (1959).
[2] Abinanti, F. R., and Huebner, R. J., *Virology*, **8**, 391 (1959).
[3] Andrewes, C. H., Bang, F. B., Chanock, R. M., and Zhdanov, V. M., *Virology*, **8**, 129 (1959).
[4] Chanock, R. M., Parrott, R. H., Cook, K., Andrews, B. E., Bell, J. A., Reichelderfer, T., Kapikian, A. Z., Mastrota, F. M., and Huebner, R. J., *New Engl. J. Med.*, **258**, 207 (1958).
[5] Hedström, H., and Isaksson, A., *Cornell Vet.*, **41**, 251 (1951).
[6] Nystedt, H., personal communication (1959).
[7] Bakos, K., Isaksson, A., and Nystedt, H., unpublished data (1958–59).
[8] Shelokov, A., Vogel, J. E., and Chi, L., *Proc. Soc. Exp. Biol. Med.*, **97**, 802 (1958).

HISTOLOGY

Preparation of Ultra-Thin Sections of Mineralized Human Enamel

THE preparation of ultra-thin sections of hard substances for electron microscopy without preliminary softening presents special difficulties. The use of industrial diamonds fashioned into knives suitable for cutting hard materials was first described by Fernández-Morán[1]. These knives, prepared with a cutting bevel of approximately 50–52°, can be used to cut or cleave thin sections of bone or tooth dentine. The sectioning of hard brittle materials, however, increases the risk of damaging the knife edge from glancing blows, since diamond is itself extremely brittle. The smallest natural cleavage angle obtainable from a diamond depends on the angles formed by the natural crystal planes (this angle is in the region of 70°), consequently in order to construct a diamond knife with the greatest possible edge strength, the two faces of the knife should form this angle. Two diamond knives both with the cutting faces diverging from the knife edge at an angle of 70° were prepared for us several years ago in the Johannesburg research laboratory of the Industrial Distributors, Ltd., under the

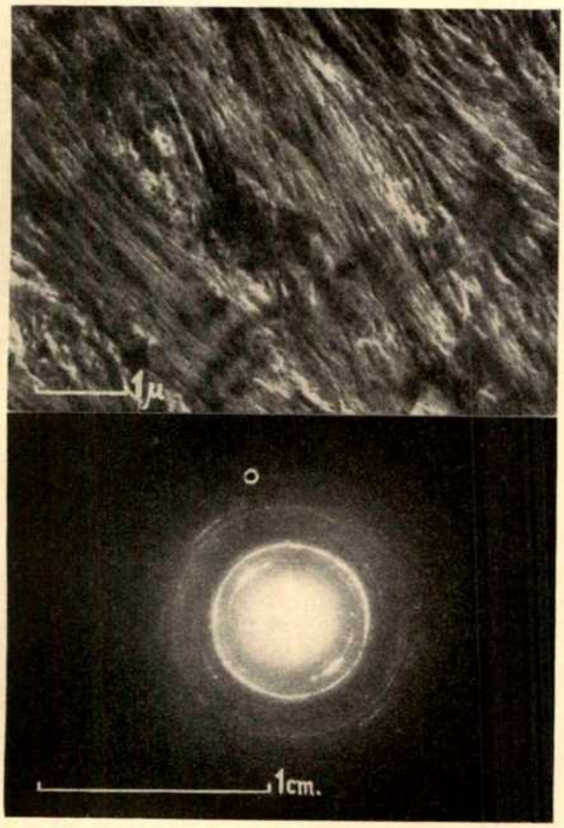

Fig. 1. Above, electron micrograph of fully mineralized unembedded human tooth enamel ; below, selected area electron diffraction pattern from 1a. The pattern is characteristic of orientated apatite crystals. Kv. 100, pole piece 2, diffraction aperture 30 μ. Instrument constant $K = 1{\cdot}93$

direction of Dr. J. F. H. Custers and the late Dr. Paul Grodzinski.

These knives have now been under test for approximately five years without repolishing, and have successfully cut human tooth enamel without detectable damage to the edge. Fully mineralized human enamel is much harder than bone or dentine and has approximately the same degree of hardness as topaz. According to Gustafson and Kling[2] the Vickers micro-hardness number of enamel varies from 300 to 400 kgm./mm.^{2}. The Vickers micro-hardness value obtained from the specimen illustrated in this communication was 412 kgm./mm.^{2}. Because tooth enamel is extremely brittle, unsupported sections disintegrate when they come into contact with the collecting fluid. They may be collected, however, by first coating the surface to be cut with a thin film of 0·2 per cent 'Formvar' dissolved in chloroform. The surface is further supported by pressing a small piece of 'Sellotape' on to the surface to be cut. By holding the 'Sellotape' the section can be removed from the upper surface of the knife at the end of the cutting stroke. The specimen is then pressed face down on to a glass slide in a drop of 'Formvar'. When dry, the 'Sellotape' can be removed by dissolving the adhesive in petroleum ether.

Thick sections mounted in this way are suitable for phase-contrast microscopy. Alternatively, after removal from the knife the section may be floated 'Sellotape' side down in petroleum ether. When the adhesive dissolves, the 'Sellotape' backing sinks leaving the section supported by 'Formvar' floating; this may then be collected on an electron microscope grid. An example of an electron micrograph and an electron diffraction pattern obtained from a section of human enamel prepared in this way is shown in Fig. 1. It is possible that this technique might be usefully applied to other brittle substances.

The preparations were examined with a Siemens Elmiskop 1 electron microscope, generously provided by the Wellcome Trust. The work was further assisted by a grant from the Medical Research Council.

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[1] Fernández-Morán, H., *Exp. Cell. Res.*, **5**, No. 1 (1953).

[2] Gustafson, G., and Kling, Ö., *Odont. T.*, **56**, 23 (1948).

RADIOBIOLOGY

Chelating Agents in the Treatment of Poisoning by Polymerizable Radioelements

The rate of reaction between a chelating agent and a hydrolysable polyvalent metal ion such as thorium (IV) is dependent on the degree of polymerization of the metal ion. It would be anticipated, therefore, that the degree to which a chelating agent might enhance the excretion rate of certain radioelements from the body would be variable even though factors such as dose and route of administration were kept constant. Accordingly, we investigated the effect of a strong chelating agent on the excretion of thorium-234 which was injected intravenously into rats in two different forms—a monomeric, diffusible form and a polymeric or colloidal form. The chelating agent used was diethylenetriaminepentaacetic acid, which forms an exceptionally strong chelate with thorium[1] (the 1 : 1 chelate has a $\log K = > 27$).

A chemical demonstration of the difference in reactivity of the two forms of thorium toward the same chelating agent is easily given. For example, when a solution of thorium nitrate ($p\text{H} = 2{\cdot}5$) is mixed with excess diethylenetriaminepentaacetic acid and the pH of the solution is increased to 7 with sodium hydroxide, the resulting solution remains clear because of the formation of the stable thorium–diethylenetriaminepentaacetic acid chelate. However, if an identical solution of thorium nitrate is neutralized before the diethylenetriaminepentaacetic acid is added, a precipitate forms which does not dissolve upon the addition of diethylenetriaminepentaacetic acid. Prolonged contact between the thorium precipitate—presumably an inorganic hydroxylated polymer[2]—and diethylenetriaminepentaacetic acid causes a slow dissolution of the precipitate over a period of many days.

In the metabolic experiments, two kinds of thorium solutions were used. The monomeric, or 'ionic', solution was prepared by adjusting a solution of carrier-free thorium-234 chloride with trisodium citrate and sodium hydroxide so that it contained 1 per cent citrate ion and had a final pH of 5·0. The polymeric solution was prepared with thorium-234 nitrate and stable thorium nitrate, adjusted with sodium hydroxide to pH 2·5 and a concentration of 1 mgm./ml. as carrier thorium. (The total concen-

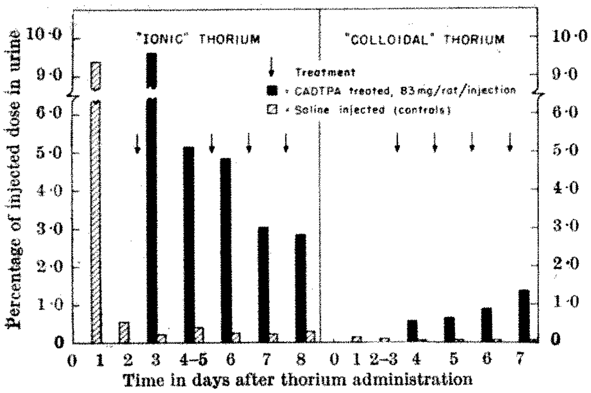

Fig. 1. Effect of diethylenetriaminepentaacetic acid (CADTPA) on the urinary excretion of thorium in rats

tration of thorium in this solution therefore was 2×10^7 times as high as in the ionic solution.) This solution gave a well-defined Tyndall beam without evidence of visible precipitation. Mature Sprague–Dawley female rats, three rats per group, injected intravenously with 0·5 ml. of one of the two solutions at a level of 0·6 μc. per rat, were placed in individual metabolism cages. Two or more days later, the treated groups received a series of four daily intraperitoneal injections of 300 mgm./kgm. of the calcium chelate of diethylenetriaminepentaacetic acid, and the controls an equivalent volume of saline. A 24-hr. urine sample was collected daily throughout the experiment for each rat. The animals were killed by cervical fracture 24 hr. after the last treatment injection. The soft tissues and femurs were wet-ashed and counted for beta activity using an end-window Geiger–Müller counter.

The results show that diethylenetriaminepentaacetic acid caused a marked reduction of the body burden of thorium only when thorium was administered in the monomeric, or 'ionic', form[3]. Under these conditions, the thorium was deposited mainly in the skeleton. The total urinary excretion of thorium for the treated groups was 34 per cent of the injected dose as compared with 12 per cent for the saline controls. The pattern of this excretion is shown in Fig. 1. The first injection of diethylenetriaminepentaacetic acid increased the 24-hr. excretion from a control level of 0·5 per cent to more than 9 per cent of the injected dose. Subsequent injections effected significant increases, but the absolute amounts which were removed steadily diminished with each injection.

The effect of diethylenetriaminepentaacetic acid treatment on rats to which thorium had been given in polymeric, or 'colloidal', form is in marked contrast to this. The total urinary excretion was only 3·6 per cent of the injected dose in the treated group as compared with 0·2 per cent in the saline controls. The thorium was deposited mainly in the liver. In addition, the pattern of the excretion is quite different, as is shown in Fig. 1. Each subsequent injection of diethylenetriaminepentaacetic acid increased the urinary excretion of colloidal thorium to a level higher than that obtained with the previous injection.

It is unlikely that these differences in the distribution and excretion of the two solutions of thorium are a function of the difference in their concentration. It has been shown that thorium is closely related to plutonium in its metabolism[4] and that the distribution and excretion of plutonium are independent of dose[5]. Rather, the results shown in Fig. 1 can be explained as follows: when 'ionic' thorium is injected it is prevented from polymerizing because of rapid complex formation with body proteins. The administration of diethylenetriaminepentaacetic acid readily removes the bound but monomeric thorium by the formation of a diffusible chelate. Each subsequent injection of diethylenetriaminepentaacetic acid removes the labile, available thorium, but the amount of thorium available for removal diminishes with each treatment. However, in the case of 'colloidal' thorium, an insoluble polymer is deposited in the tissues. In the presence of sustained levels of diethylenetriaminepentaacetic acid within the tissues[6] the particles of thorium become slowly depolymerized. Consequently the amount of labile thorium, that is, thorium–diethylenetriaminepentaacetic acid chelate, increases with time so that subsequent injections of chelating agent result in an increase of excretion over the previous injection.

Details of these and other metabolic experiments involving the removal of thorium with chelating agents will be published elsewhere. The results cited here emphasize the point that the success of the use of chelating agents in the treatment of radioelement poisoning is dependent on the chemical and physical properties of the radioelement.

This work was performed under the auspices of the U.S. Atomic Energy Commission. We are indebted to Miss Elizabeth Moretti for technical assistance.

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[1] Boguicki, R. F., and Martell, A. E., *J. Amer. Chem. Soc.*, **80**, 4170 (1958).
[2] Hietanen, S., *Acta Chem. Scand.*, **8**, 1626 (1954).
[3] Fried, J. F., Lindenbaum, A., and Schubert, J., *Fed. Proc.*, **18**, 905 (1959).
[4] Boone, I. V., Rogers, B. S., White, D. C., and Harris, P. S., *Amer. Indust. Hyg. Assoc. J.*, **19**, 285 (1958).
[5] Weeks, M. H., Oakley, W. D., and Thompson, R. C., *Rad. Res.*, **2**, 237 (1955).
[6] Schubert, J., and Lindenbaum, A. (submitted for publication).

Atmospheric Radiocarbon Activity in 1959

ATTENTION has already been directed to the appreciable rise in the atmospheric radiocarbon activity following the testing of nuclear weapons[1–4]. On the basis of data obtained up till 1958, Broecker and Walton[1] estimated a rise in the atmospheric radiocarbon concentration in the northern hemisphere of 5 per cent per annum, though Münnich and Vogel[2] had arrived at a rate of rise of 3·2 per cent per annum. Measurements made in the summer of 1959, however, indicate a general level of activity greatly in excess of what might have been expected from either of these estimations.

In an attempt to evaluate the progressive rise in concentration in Britain over the past seven years, samples of annual oat crops were kindly supplied by the National Institute of Agricultural Botany, Cambridge, from its seed collection, and assayed for radiocarbon in this Laboratory. Oats have a very short growing season, and photosynthesize principally at the time of year when Münnich and Vogel[5] have demonstrated that the seasonal intensity of radiocarbon has reached its maximum. This variation has been ascribed to the rise in the tropopause in

Table 1

Sample No.	Year of harvest	Change from 1953 (per cent)
Q–436	1953	0·0 ± 0·5
Q–437	1954	− 0·7 ± 0·5
Q–438	1955	+ 1·2 ± 0·5
Q–439	1956	+ 3·0 ± 0·5
Q–440	1957	+ 11·5 ± 0·6
Q–441	1958	+ 14·5 ± 0·6
Q–442	1959	+ 31·8 ± 0·7

summer, allowing part of the stratosphere to mix freely with the troposphere. The crops were grown in and around Cambridge, and thus the measurements from year to year are directly comparable.

If the level of activity of 1953, before any appreciable nuclear weapon testing had been carried out, is taken as the standard activity for subsequent years, the results (Table 1) may be expressed as a percentage variation from this value.

The large difference in activity between the summers of 1958 and 1959 must be attributed to the intense testing programmes undertaken late in 1958, and its delay in appearing in the atmosphere probably lies in the mechanism of the seasonal variation already referred to.

It is interesting to note that the activity of a sample of human neck hair cut in the late summer of 1959 was 16·5 per cent greater than the 1953 oat sample, and is thus considerably less than the activity of the 1959 oat crop. During the period of growth of this hair we can assume that the population was in the main still consuming the cereal and root crops of the previous season.

If the present suspension of nuclear weapon tests were to become permanent, the future level of the atmospheric radiocarbon activity, together with that of the surface ocean water, should furnish further valuable information about the exchange rates of carbon dioxide between the various phases of the radiocarbon reservoir[1,2].

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[1] Broecker, W. S., and Walton, A., *Science*, 130, 309 (1959).

[2] Munnich, K. O., and Vogel, J. C., *Naturwiss.*, 14, 327 (1958).

[3] de Vries, Hl., *Science*, 128, 250 (1958).

[4] Rafter, T. A., and Ferguson, G. J., *N.Z. J. Sci. and Tech.*, B, 38, 871 (1957).

[5] Munnich, K. O., and Vogel, J. C., in Godwin, H., *Nature*, 184, 1365 (1959).

BIOLOGY

Implantation of Tissue Transplants in the Uteri of Pseudopregnant Mice

It was thought that transplants of tumour into the uterine cavities of pseudopregnant mice might simulate the invasive properties of the embryonic trophoblast sufficiently to throw light on the uterine reactions involved in the implantation of the embryo. Since the initial results of these studies, which are still in progress, appear to show clearly that the site of implantation is determined by the uterus, a preliminary report may be justified.

Table 1. TUMOUR TRANSPLANTS IN PSEUDOPREGNANT LAB. GREY AND C57 BLACK MICE

Day of transplantation (after mating)	No. of experiments	No. showing implantation of tumour
0	4	0
1	6	0
2	6	(1*)
3	8	0
4	14	12
5	8	1
6	5	2 (+ 1*)
7	4	0
8	5	0
9	4	0
Diœstrus	3	0

* Animals in which only a few melanoma cells have invaded the mucosa.

Mature females of strains *C*57 Black (pure line) and Lab. Grey (stock) mated with vasectomized males were used. The day on which the vaginal plug was found was taken as day 0 in the pseudopregnant cycle. The tumour used was the Harding–Passey[1] melanoma. Laparotomies were performed under ether anæsthesia and the tissue fragment inserted into the uterine lumen with a Bashford implantation needle (bore 0·625 mm.), ligatures being placed around the uterus above and below the transplant, which otherwise was expelled either through the incision or the cervix. The tumour was invariably transplanted into the right uterine horn and usually a fragment of the animal's own body-wall muscle into the other horn. Transplantations were performed from days 0 to 9 and the animals killed 2 days later. The uteri were then removed, fixed in Zenker's fluid and sectioned transversely. No differences were observed between the *C*57 Black and the Lab. Grey strains.

It is apparent from Table 1 that implantation of the tumour in the uterine mucosa within the 2-day duration of the experiments was most frequent when the transplantation was effected on the fourth day of pseudopregnancy. Implantation was not observed after transplantation on days 0–2 or 7–9 of pseudopregnancy or during diœstrus. The autografts of muscle in the other uterine horn were made only

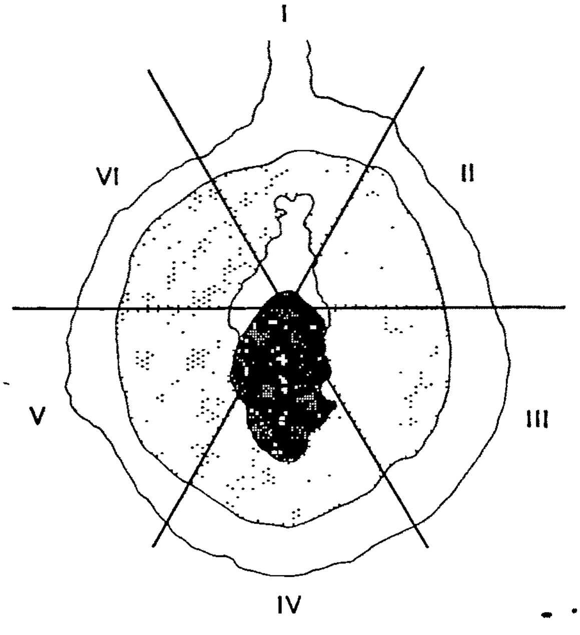

Fig. 1. Camera lucida outline diagram of tumour fragment (cross-hatched) implanting antimesometrially in the uterine mucosa (stippled) of a pseudopregnant mouse. The sectors employed in Tables 2 and 3 for defining the position of attachment are superimposed

Table 2. UNIFORMITY OF ANTIMESOMETRIAL IMPLANTATION OF THE TUMOUR IN LAB. GREY AND *C57* BLACK MICE

	Experiment No.	Day of transplantation	Sectors of uterus containing implant					
			I	II	III	IV	V	VI
Lab. Grey	1	4			+	++	+	
	2	4			+	++	+	
	3	4				++	+	
	4	4			+	++	+	
	5	4			+	++	+	
	6	4			+	++		
	7	4			++	+		
	8	4			+	++	+	
	9	4			+	++		
	10	5				++		
	11	6				+	+	
C57	12	4			++	+		
	13	4			+	+		
	14	4			+	++		
	15	6				+	++	

++, Implantation extending across whole sector; +, implantation spreading into sector.

Table 3. UNIFORMITY OF ANTIMESOMETRIAL IMPLANTATION OF MUSCLE TISSUE IN LAB. GREY AND *C57* BLACK MICE

	Experiment No.	Day of transplantation	Sector of uterus containing implant					
			I	II	III	IV	V	VI
Lab. Grey	—*	3			++	+		
	—*	3			+	++	+	
	1	4			+	++	+	
	2	4			+	++	+	
	3	4			+	++	+	
	4	4				++	+	
	—*	4			+	++	++	
	—*	5				+	++	
	—*	5					++	+
C57	12	4			+	++		
	13	4				++	+	
	14	4			+	++	+	
	—*	5				++	++	+
	15	6			+	++		

—*, Experiments where the tumour tissue did not become implanted, but the control muscle tissue did.

on days 3–6 of pseudopregnancy and failed to become implanted in only one instance in fifteen experiments.

The melanoma or muscle tissue when it became implanted did so invariably on the antimesometrial side of the lumen as does the normal embryo in the mouse. As a rule the tumour had penetrated the epithelium and deeply invaded the mucosa, the camera lucida outline diagram of a section in Fig. 1 being typical. The muscle tissue became attached to the mucosa, usually with destruction of the epithelium but without further invasion of the mucosa. The position and extent of the attachments are represented in Tables 2 and 3 by dividing the camera lucida outline drawings of the median transverse section into 6 sectors, as in Fig. 1, sector I being mesometrial and sector IV antimesometrial.

The tissue fragments implanted were many times larger than a normal embryo, being at least five times the diameter, yet whether homografts of melanoma or autografts of muscle were employed, implantation occurred with remarkable uniformity antimesometrially in precisely the region where an embryo would have become implanted. It is difficult to escape the conclusion that the antimesometrial site of implantation is determined by the uterus and that the properties of the mucosa at the appropriate stage of pseudopregnancy are such as to encourage the attachment of all these diverse tissues. The work of McLaren and Michie[2] has shown that the siting of implantations along the length of the uterus is not predetermined.

Other aspects of this work still in progress, such as the induction of the decidual reaction, together with a full report of the histological findings, will be published later.

I am indebted to Prof. F. W. R. Brambell for his advice and encouragement, to Prof. A. Haddow, of the Chester Beatty Institute, for supplying the tumour, and to the staff of this Department for technical assistance. The work was performed during the tenure of a Research Studentship of the Department of Scientific and Industrial Research.

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[1] Harding, H. E., and Passey, R. D., *J. Path. Bact.*, 33, 417 (1930).
[2] McLaren, A., and Michie, D., Mem. Soc. Endocrinol. No. 6, 65 (1959).

Effect of Soil Drying on the Viability of *Heterodera major* Cysts

WINSLOW[1] has shown that dried cysts of *Heterodera major* do not hatch well. He found that more than two thousand larvæ hatched from cysts recovered from wet soil, whereas only seven larvæ hatched from cysts which had been recovered from dried soil. Hesling[2], in an experiment carried out at Rothamsted, confirmed Winslow's results, and concluded that this eelworm is quite sensitive to drying.

Cysts of *H. major* were found on the roots of volunteer oat plants which were growing in discarded infected soil. Prior to being discarded, this infected soil had been air-dried in the laboratory and, since cysts were found on the roots of oat plants growing in this soil later, it would appear that the drying of the soil did not seriously affect the viability of the cysts in it.

Early in January 1955, a quantity of infected soil was placed in a heap and was thoroughly mixed to ensure uniformity in cyst-distribution throughout. A microplot (in the field) was filled with this mixture and at certain intervals over a period of fifteen months a portion of soil, sufficiently big to fill ten test-tubes, was taken from the microplot and was air-dried. When the soil was dry, the ten test-tubes were filled with it, and it was thus stored in this air-dried condition in the laboratory until sowing time. This gave lots of infected soil which had been air-dried for periods of 1, 3, 6, 9, 12 and 15 months. Ten tubes containing infected soil which was not air-dried and ten tubes containing non-infected soil were included in the experiment, also, and were used as controls.

On April 16, 1956, all tubes were sown with oats (variety Sun II), and were plunged in a bed of peat-moss in the field.

By the middle of June, cysts of *Heterodera major* were found to be quite plentiful on the roots of plants growing in the infected soil which had been air-dried for periods of one and three months and in the infected soil which had not been air-dried. It must be emphasized that the soil, when dried for these periods, was 'dust-dry', and was certainly dry enough for cyst extraction using the Fenwick apparatus.

Despite repeated examinations, cysts were not observed on the roots of plants growing in infected soil which had been air-dried for periods of 6, 9, 12 and 15 months. It was concluded that, if infected soil is air-dried, and kept in an air-dried condition for a period of 6 months or longer, the cysts lose their viability and their contents become incapable of hatching and invading the roots of a host-plant.

It is interesting to record that the effect of drying on the viability of the cysts was reflected in the growth of the plants. The plants growing in soil which had been air-dried for periods of 6, 9, 12 and 15 months were comparable in size and vigour with plants which were growing in tubes of non-infected soil, and showed no symptoms of eelworm attack. On the other hand, plants growing in the infected soil which had been air-dried for periods of 1 and 3 months showed symptoms of severe eelworm attack and were very dwarfed and diseased-looking.

The hatching experiments of Winslow[1] and of Hesling[2] suggest that cysts of *H. major* are far more sensitive to drying than the above-mentioned observations made by me found them to be. It is possible that the effect of drying becomes more pronounced when the cysts are removed from the soil, a factor which might explain this apparent contradiction in results.

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[1] Winslow, R. D., *Ann. App. Biol.*, 43, 19 (1955).

[2] Hesling, J. J., *Nematol.*, 1 (1), 56 (1956).

Albinism in *Cryptomys hottentotus* Lesson, 1826

In an earlier communication[1] I reported the occurrence of what appeared to be an albino specimen of the above species of mole-rat. I am grateful to Miss D. Watson for a further and similar example, collected on November 12, 1958, at a point within a few hundred yards of where the first animal was taken.

As in the previous instance the mammal was taken on the surface, and was examined while still alive. In all respects the coloration was similar to that previously described, except that the dietetic staining was not so pronounced. The specimen, an adult male, was preserved entire, and revealed the following body measurements: head and body, 130·0 mm.; tail, 15·0 mm.; hind foot s.u., 22·0 mm. The cheek teeth were again simple, 4/4.

Previously I stated that normal specimens of *C. hottentotus* at Inyanga possessed a "variable white head-spot which is sometimes vestigial". Since this was written I have handled specimens of adults, taken side-by-side with head-spotted examples, in which the head-spot has been entirely absent. Thus it seems that Inyanga specimens are the antithesis of those described by Roberts[2], who notes the position regarding head-spots as "white frontal spots are not infrequent, but abnormal". The converse may be said to be true at Inyanga.

It might be of further interest to note that while the tail normally has a length of about 11·0–22·0 per cent of head and body-length, some specimens may have tails as long as 30·0 per cent of head- and body-measurement.

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[1] Turnbull-Kemp, P. St. J., *Nature*, 182, 63 (1958).

[2] Roberts, A., "The Mammals of South Africa" (Trustees of "The Mammals of South Africa" Book Fund, Johannesburg, 1951).

Quantitative Immuno-Analysis in Gel Plates

I cannot let pass unchallenged certain assertions made by Wright[1] in his communication in *Nature* of May 2, 1959, with reference to my method for quantitative immuno-analysis in agar gel plates[2]. Dr. Wright states: "Although there are several methods available, none of them is easily applicable to a multi-antigenic system, and in addition the concentrations of the proteins can only be estimated separately, even under favourable conditions, at ±10 per cent and more usually ±50 per cent".

The entire joy of the quantitative plate method, as outlined in my original communication[2] and since extended[3-5], is that it permits the ready titration of multi-antigenic systems and the simultaneous assay of individual antigens, often on a single plate. It has the further advantage of not requiring titration to an end-point, assays being readily performed by comparison of precipitation patterns.

As for the sensitivity of the technique, claims below ±10 per cent have not been made simply because we have never felt the need for greater accuracy and have been content to use, at the narrowest, 1·1 × serial dilutions. A narrower dilution gradient, as used by Wright in his technique, could well produce accuracies better than ±10 per cent.

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[1] Wright, S. T. C., *Nature*, 183, 1282 (1959).

[2] Feinberg, J. G., *Nature*, 177, 530 (1956).

[3] Feinberg, J. G., *Int. Arch. Allergy*, 11, 129 (1957).

[4] Feinberg, J. G., Proc. Fourth Int. Congr. Biol. Standards, 194 (1958).

[5] Feinberg, J. G., *Immunol.* (in the press).

MICROBIOLOGY

Exogenous Respiration in *Nitrobacter*

During the course of an investigation of the oxidation of nitrite by the nitrite-oxidizing bacterium, *Nitrobacter*, it was noted that the addition of sodium nitrite but not sodium nitrate to heavy cell suspensions caused the immediate appearance of reduced cytochrome bands at 590, 551 and 513 mμ when viewed in the hand spectroscope[1]. This phenomenon, which has been noted previously by Lees and Simpson[2], is due to the passage of electrons through the respiratory chain when nitrite is oxidized to nitrate. Whether or not nitrifying bacteria respire in response to substrates other than their energy source has been disputed, although Bömeke[3] reported a slight stimulation of the respiration of *Nitrobacter* when certain metabolites were added to cell suspensions in the Warburg respirometer. If some reduced substance is oxidized by resting cells and if a cytochrome-linked pathway is employed for the oxidation of all substances which can permeate the cell, then any substance which might increase the rate of respiration in the Warburg respirometer should also cause a reduction of the cellular cytochromes. The validity of this rationale was tested in the following manner.

Cultures grown for 4–8 days at 25° C. on a reciprocating shaker in a mineral medium (gm./l.: sodium nitrite, 1·0; sodium chloride, 0·5; dihydrogen potassium phosphate, 0·44; dipotassium hydrogen phosphate, 2·88; crystalline magnesium sulphate heptahydrate, 0·3; ferrous sulphate, 0·003; sodium carbonate, 1·0; pH to 7·0–7·2) were harvested by centrifugation and washed until free of nitrite. Heavy cell 'creams' were thoroughly aerated for 30–60 min. and then observed in the Hartridge reversion spectroscope (slit-width 125μ). The spectrum obtained was that of an oxidized cytochrome system, and it remained oxidized for at least 30 min. after the cessation of aeration. Various substrates (0·1 ml. of neutralized 0·05 *M* solutions) were pipetted into the cell suspension and it was observed for 30 min. for spectral changes. As shown in Table 1, only sodium nitrite and formate caused a reduction of the cellular cytochromes. The bands appeared immediately (within 10 sec.) and were in identical positions for both substances.

Table 1. THE EFFECT OF SELECTED METABOLITES ON THE RESPIRATION OF *Nitrobacter*

Addition	Reduced cytochrome bands appearing, mμ	Q_{O_2} (28° C.)
None	None	2·4
Sodium nitrite	590, 551, 513	51·7*
Sodium nitrate ..	None	N.T.
Sodium formate ..	590, 551, 513	10·8*
Sodium acetate ..	None	4·8†
Sodium lactate ..	None	2·2
Glucose	None	1·8
Sodium citrate .	None	1·3

N.T. = Not tested.

* No lag.

† After 50-min. lag during which respiration was equivalent to endogenous.

The effect of these substances on the oxygen uptake of washed cells was tested by conventional techniques in the Warburg respirometer (final concentrations: substrate, 25 μ*M*; pH 7·2 phosphate buffer, 400 μ*M*; potassium hydroxide in the centre well). Oxygen uptake was followed for a total of 2 hr. after tipping in the substrate, although nitrite was completely oxidized within the first 45 min. It may be noted in Table 1 that of the substances tested, only formate and nitrite readily stimulated oxygen uptake. Thus formate is a metabolizable substrate for intact *Nitrobacter* cells. Whether the slight increase in oxygen uptake with acetate is significant is not known—however, it lies within the range of experimental error. Thus only those substances which evoked the rapid reduction of cellular cytochromes caused an increase in uptake of oxygen. The spectroscopic method, therefore, appears to offer a rapid and convenient way to survey potential substrates for *Nitrobacter*.

These preliminary results suggest that the following interesting speculations be tested: The autotroph, *Nitrobacter*, differs from better-known heterotrophic bacteria in that it is not permeable to many of the usual metabolic intermediates. This is reflected by the lack of effect of common metabolites on respiration. It is reasonable to postulate that many of the common enzyme systems are operative within the cell, but the use of intact cells gives no evidence of this potentiality due to the impermeability to the required substrate. Cell-free preparations, however, should possess many of the enzyme systems which catalyse the utilization of some of the substances listed in Table 1. These hypotheses are now being tested experimentally by spectroscopic and manometric techniques.

This work was aided by a grant from the Society of the Sigma Xi and a contract between the Office of Naval Research, Department of the Navy, and the University of Florida, No. 103–431 (Nonr–2651 (00)).

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[1] Silver, W. S., and Watt, C. V., *Bact. Proc.*, **24** (1959).

[2] Lees, H., and Simpson, J. R., *Biochem. J.*, 65, 297 (1957).

[3] Bomeke, H., *Arch. Mikrobiol.*, 10, 385 (1939).

Shape of Ultra-violet Inactivation Curves of Transforming Deoxyribonucleic Acid

IN analysing the effects of radiation on biological systems, it is customary to plot the logarithm of the surviving fraction (or of the fraction of the biological activity remaining) against the applied dose. When such a plot gives a straight line passing through the origin, the radiation sensitivity of the system may be characterized by a single constant, which is conveniently taken as the negative slope of the logarithmic inactivation curve. Such a curve is consistent with the assumption that the inactivation results from independent, single events (inactivating 'hits') occurring at random[1].

When the inactivation of bacterial transforming deoxyribonucleic acid by ionizing or ultra-violet radiation is analysed by this procedure, the curves are not straight lines, but bend steadily in the direction of shallower slopes as the radiation dose increases[2-4]. 'Multicomponent' curves of this character require two or more constants for their characterization. They are obtained if the irradiated population is heterogeneous in its sensitivity to radiation[1], each class being inactivated according to a straight line curve (exponential law). In such cases the plot becomes a straight line at high doses where the survivors consist chiefly of members of the most resistant class. These curves can also result from suitable heterogeneity in the substructure of each individual. Deoxyribonucleic acid inactivation results for ionizing radiations have usually been interpreted in terms of one of these schemes, with attempts to determine the 'limiting slope' at high radiation doses corresponding to one resistant class of deoxyribonucleic acid molecules or one class of molecular substructures[5,6].

In semi-logarithmic inactivation curves for *Hemophilus influenzae* transforming deoxyribonucleic acid exposed to 254 mμ ultra-violet radiation, we find a steadily decreasing curvature at high doses[4], in agreement with other workers[3]. These inactivation curves, for all the genetic markers tested, can be converted into straight lines over a wide range of high doses by plotting the logarithm of the transforming activity against the *logarithm* of the ultra-violet dose (instead of against the ultra-violet dose itself) (unpublished work of S. H. G.). Such a relation on a log–log plot is incompatible with a straight line on a semi-log plot, and indicates a power-law inactivation at these doses rather than some variety of exponential law.

Preparation, ultra-violet inactivation and assay of transforming deoxyribonucleic acid were carried out as detailed in ref. 7 (except that the concentration of deoxyribonucleic acid in contact with bacterial

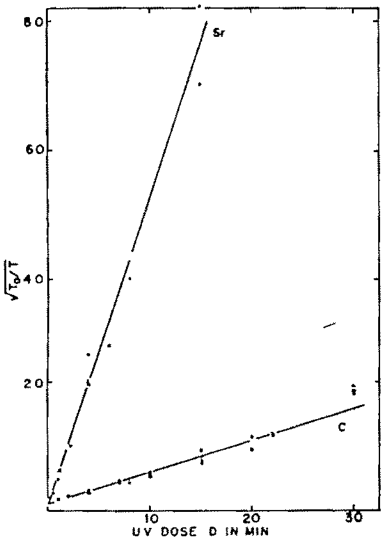

Fig. 1. Reciprocal square root of the fractional transforming activity $\sqrt{(T_0/T)}$ vs. the radiation dose D (in min. at 1.650 ergs mm.$^{-2}$ min.$^{-1}$ of 254 mμ radiation). T_0 is the transforming activity before irradiation, and T the activity afterwards of the genetic markers, streptomycin resistance (Sr 2 000 γ/ml.) and cathomycin resistance (C 2·5 γ/ml.). Deoxyribonucleic acid concentrations in contact with cells during transformation were as follows: (●) 5×10^{-3} γ/ml.; (○) 5×10^{-2} γ/ml.; (×) 10^{-1} γ/ml.

cells during transformation was as indicated in the caption of Fig. 1). The ultra-violet dose-rate in ergs/mm.^{2}/min. was estimated from the rate of inactivation of $T2$ bacteriophage[8]. For all markers tested, a plot of the reciprocal square root of the transforming activity against dose leads to straight lines over the entire dose-range (Fig. 1), implying an inverse square law:

$$T/T_0 = 1/(1+CD)^2 \qquad (1)$$

Here T_0 is the initial transforming activity of a preparation, T is the activity after exposure to a radiation dose D, and C is a constant depending on the particular genetic marker under study. This single constant, which may be used to characterize the sensitivity to radiation of the marker, is equal to minus one-half the initial slope of a semi-logarithmic plot of the same results. The entire curve may be used, however, in determining it, instead of only the initial portion. From the slopes of the lines in Fig. 1, the relative sensitivities to ultra-violet radiation of the streptomycin-resistance and cathomycin-resistance markers of *H. influenzae* are easily specified as being in the ratio of 10 to 1, a fact which would be difficult to state quantitatively from the usual presentation. Genetic markers for erythromycin resistance and viomycin resistance (which also give straight lines on an inverse square root plot) have sensitivities very close to that of the cathomycin marker.

The straight line region, of slope minus 2, which appears on a log–log plot at high doses, corresponds to the entire dose-range in which $CD \gg 1$; that is, to the region in which the transforming activity is less than about 10 per cent of that preceding irradiation.

The absoluteness with which relation (1) holds can be determined only by extensive experience. Experiments covering the low dose-range more thoroughly than those reported here show that it describes inactivation very well in this region. At the very highest doses it may perhaps predict a slower inactivation than is actually found by experiment, since the points shown for cathomycin resistance at 25 and 30 min. (41,000 and 50,000 ergs/mm.^{2}) lie slightly off the theoretical fitted curve. This discrepancy is not clearly present in all experiments and its reality is not certain.

At present we offer no theoretical interpretation of relation (1), which is entirely empirical. Its simplicity suggests that the complex ultra-violet inactivation curves observed on semi-logarithmic plots may, after all, arise from a relatively simple mechanism, although one which possibly differs from the type of inactivation envisioned by elementary target theory. The simpler and apparently more appropriate manner of presenting ultra-violet inactivation data which now becomes available should facilitate studies aimed at determining this mechanism.

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[1] Lea, D. E., "Actions of Radiation on Living Cells", 72 (Macmillan Co., New York, 1957).

[2] Ephrussi-Taylor, H., and Latarjet, R., *Biochim. et Biophys. Acta*, 16, 183 (1955).

[3] Lerman, L. S., and Tolmach, L. J., *Biochim. et Biophys. Acta*, 33, 371 (1959).

[4] Rupert, C. S., Goodgal, S. H., and Herriott, R. M., Proc. of First Nat. Biophysics Conf., H. Quastler and H. J. Morowitz, edit., 219 (Yale Univ. Press, New Haven, 1959).

[5] Guild, W. R., and deFilippes, F. M., *Biochim. et Biophys. Acta*, 26, 241 (1957).

[6] Ephrussi-Taylor, H., in "The Chemical Basis of Heredity", 299, edit. McElroy and Glass (Johns Hopkins Press, Baltimore, 1957).

[7] Rupert, C. S., Goodgal, S. H., and Herriott, R. M., *J. Gen. Physiol.*, 41, 451 (1958).

[8] Latarjet, R., Morenne, P., and Berger, R., *Ann. Inst. Pasteur*, 85, 174 (1953).

STATISTICS

Performance of a Stochastic Net

Stochastic nets have been proposed by several authors[1–3] as models of cognitive activity. So far, these have been dominated by heuristic constraints designed to show some analogy with the nervous system or merely for ease of computation. An adequate mathematical theory has, as was recognized by Uttley[4], to show how such a device can learn relations.

In the following I give a model which with a simple and natural constraint has the ability to detect linear relations and record their components in a principal axis frame of reference; as such it has obvious relations with well-known topics of linear analyses, namely, multiple factor[5], and prediction and filtering[6] theory.

Consider a collection of n cells i; let α_i, α^i, be their inputs and outputs respectively and W_{ij} the strength of connexion from cell j to i such that if α_i^S is the input from sensory field S to cell i, we have: $\alpha_i = \alpha_i^S + W_{ij}\,\alpha^j$ (summation over repeated indices), assuming unit linear amplification for each cell i (that is, $\alpha^j = \delta^{ji}\alpha_i$); the effective amplification with positive feedback W_{ii} is $(1-W_{ii})^{-1}$ so that the above can be written:

$$\alpha_i^S = (\delta_{ij} - W_{ij})\,\alpha^j = A_{ij}\alpha^j \text{ say} \qquad (1)$$

W_{ij}, α_i^S may be positive or negative according as their synaptic endings are excitatory or inhibitory.

Consider n outputs of receptors from a sensory field S such that the covariance:

$$C_{ij} = \lim_{T\to\infty} C_{ij}(T) = \lim_{T\to\infty}\frac{1}{T}\int_0^T \alpha_i^S(t)\,\alpha_j^S(t)\,\mathrm{d}t \qquad (2)$$

exists. The structure of this matrix reflects the simplest properties of the field S, namely, if the rank $r<n$ then there will be $n-r$ linear relations between the α_i^S. If now we can arrange that:

$$\lim_{t\to\infty}\ \sum_k A_{ik}(t)\,A_{jk}(t) = C_{ij} \qquad (3)$$

then we shall have a description of the field $S(t)$ in terms of n outputs $\alpha^j(t)$ which are orthonormal in the sense that:

$$\lim_{T\to\infty}\ \frac{1}{T}\int_0^T \alpha_i(t)\,\alpha_j(t)\,\mathrm{d}t = \delta_{ij} \qquad (4)$$

and of which therefore $n-r$ must be arbitrary, that is, generated by the net itself. Were it not for this, which will allow us to distinguish them from the r outputs representing $S(t)$, we should have gained nothing from the transformation (1).

To realize (3), let us put:

$$t\frac{\partial A}{\partial t}ik + \frac{1}{2}A_{ik} = \frac{1}{2}\alpha_i^S\alpha_k \qquad (5)$$

Multiplying through by A_{jk}, using the symmetry of C_{ij} and summing over k it is easy to show that this satisfies (2) and (3). However, (3) only determines A_{ij} up to an orthogonal transformation corresponding to an arbitrary rotation of the orthogonal frame; the additional condition:

$${}_k\Sigma\, A_{ki}A_{kj} = \lambda_i\,\delta_{ij} \qquad (6)$$

determines the principal axis solution, where λ_i are the proper values of C_{ij}. If (6) holds, then we have:

$$A_{ij}\,\alpha_i^S = \lambda_j\,\alpha_j \qquad (7)$$

multiplying (5) by A_{ij} and summing this time over i, one can check that (6) follows.

Using (1) and (4), the corresponding condition for the connexions is:

$$t\frac{\partial W}{\partial t}ij + \frac{1}{2}W_{ij} = \frac{1}{2}(\alpha_i - \alpha_i^S)\alpha_j \qquad (8)$$

which means that the (negative) connexion between two cells is proportional to the correlation between the activity of the cell from which the connexion is coming and the stimulus on the cell to which it is going. For the feedback of a cell to itself the same is true of the reciprocal gain. This shows that if α_j is uncorrelated with any of the α_i^S, then the cell j is effectively isolated and its gain rises to an infinite value such that α_j is a self-generated signal of unit variance. Under these conditions an indefinitely flat peak in the frequency response will produce a single-frequency sinusoidal output. If no two of these frequencies are exactly equal, then (4) will certainly be satisfied. It is not unreasonable to compare this behaviour with the α-rhythm of the inattentive brain. Furthermore, the principal axes solution in which the co-ordinate axes are determined by successive extrema of the stimulus vectors is the one most indicated by physiological evidence.

An example of the application of such a net would be the following: suppose we have n audio receptors responding to a frequency band $\Delta\nu_i$, and let their (rectified and smoothed) outputs be $\alpha_i^S(t)$. For a sensory field such as normal speech, the latter will be highly correlated, so that a smaller number r will be able to convey the information. In fact, just such a procedure has been used[7] in a speech communication channel known as the 'Vocoder'. For example, with $\Delta\nu_i = 15$ c./s., $n = 16$, we can have $r = 10$ without loss of fidelity.

However, the significance of the net as a model of local brain activity would appear to be the following. If we drop the rigour implied in the infinite limit of (2), then what the net does for us is to factorize a sensory field into a part which has no constancy in time and a part which is invariant over a period of time T, and which is expressed in the values of the A_{ij}. Over longer periods of time, the latter may vary and could be the subject of analysis by a net of longer time constant. In this way knowledge is possible in spite of the conditional and incomplete nature of all relations.

Note added in proof. To obtain the principal axis solution uniquely some modification of (5) is required; it will be given elsewhere.

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[1] Neumann, J. von, "The Computer and the Brain" (Yale, 1958).
[2] Beurle, R. L., *Phil. Trans. Roy Soc*, 240, 55 (1956).
[3] Rosenblatt, F., *Psych Rev*, 65, 386 (1958).
[4] Uttley, A. M, Proc Symp. Information Theory, London (1950).
[5] Thurstone, L. L., "Multiple Factor Analyses" (Chicago, 1947).
[6] Wiener, N., "Extrapolation, Interpolation and Smoothing of Stationary Time Series" (Chapman and Hall, London, 1950).
[7] Kramer, H. P., and Mathews, M. V., *Trans. Inst. Rad. Eng*, *IT*-2, 41 (1956).

Standardized t

Students' t may be standardized by expressing it in units of its own standard deviation; thus, $t_s = t\sqrt{\{(\nu-2)/\nu\}}$, where ν is the number of degrees of freedom. The procedure reduces the variability of the tails with ν for a given probability, P, and in particular, for $P = 0{\cdot}05$ and $\infty \geqslant \nu \geqslant 4$, t_s lies between 1·96 and 2·0 whereas t lies between 1·96 and 2·776. This finding extends the applicability of certain 'large' sample methods down to 4 degrees of freedom. The standardizing is extremely simple to apply—merely divide the error sum of squares by $\nu - 2$ instead of by ν and use this value to calculate the appropriate standard error (*SE*). For example, if d is the difference between two means and $d \geqslant 2\ SE$, $P \leqslant 0{\cdot}05$, that is, the difference is significant at the 5 per cent level. If $d < 1{\cdot}96\ SE$, $P > 0{\cdot}05$ and the difference is not significant. Between these two limits of d, $P \simeq 0{\cdot}05$; actually $0{\cdot}0535 > P > 0{\cdot}0455$. Again, $\pm 2\,SE$ will give a close approximation to 95 per cent confidence limits (actual value between 95 and 95·45 per cent). When the formula is applied to the correlation coefficient, r, we find that r is significant at the 5 per cent point if $r \geqslant 2/\sqrt{(\nu+2)}$.

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FORTHCOMING EVENTS

*(Meetings marked with an asterisk * are open to the public)*

Monday, February 22

Institution of Electrical Engineers (at Savoy Place, London, W.C.2), at 5.30 p.m.—Discussion on "Are We Making the Best Use of Research Resources" opened by Mr. L. Rotherham.

Institution of Mechanical Engineers, Education Group (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Discussion on "The Place of Management Studies in the Training of an Engineer".

Monday, February 22—Friday, February 26

Engineering Materials and Design Exhibition and Conference (at Earls Court, London, S.W.5).*

Tuesday, February 23

University of London (at Queen Mary College, Mile End Road, London, E.1), at 1.30 p.m.—Dr. Donald Hunter: "Historical Perspective in Science and Medicine".

Institute of Marine Engineers (at the Memorial Building, 76 Mark Lane, London, E.C.3), at 5.30 p.m.—Mr. A. Bell: "Corrosion and Fires in Marine Boiler Air Preheaters".

Institute of Physics, Electronics Group (at 47 Belgrave Square, London, S.W.1), at 5.30 p.m.—Dr. W. M. Lomer: "Magnetic Properties of Alloys".

Institution of Chemical Engineers (at the Geological Society, Burlington House, Piccadilly, London, W.1), at 5.30 p.m.—Mr. H. E. Rose: "Vibratory Ball-mills".

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Dr. W. R. S. Doll: "Epidemiological Methods in Leukæmia Research".* (Thirteenth of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on February 25 and March 1.).

University of London (in the Chemistry Theatre, University College, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. J. A. B. Gray: "The Place of Physiology in University Education".*

Institution of Electrical Engineers, Education Discussion Circle (at Savoy Place, London, W.C.2), at 6 p.m.—Discussion on "Courses for Electrical Technicians" opened by Prof. M. W. Humphrey Davies.

Institution of Mechanical Engineers, Lubrication Group (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Discussion on "Water Lubrication".

British Interplanetary Society (joint meeting with the London Branch of the Institution of Plant Engineers, at the Royal Society of Arts, John Adam Street, Adelphi, London, W.C.2), at 7 p.m.—Mr. R. H. B. Forster and Mr. L. Breen: "Some Problems Encountered in the Design of Large Rocket Test Beds".

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Mr. A. J. Troughton: "Relationship between Theory and Practice in Structural Problems".

Society of Instrument Technology (at Manson House, 26 Portland Place, London, W.1), at 7 p.m.—Mr. D. F. White and Mr. L. E. Taylor: "Backscatter Method of Wall Thickness Measurement"; Mr. M. V. James: "Ultrasonic Resonance Method of Wall Thickness Measurement".

Wednesday, February 24

University of London (at the Postgraduate Medical School of London, Ducane Road, London, W.12), at 2 p.m.—Dr. R. R. Race: "The Genetics of Blood Groups".*

Royal Society of Arts (at John Adam Street, Adelphi, London, W.C.2), at 2.30 p.m—Prof A. C. B. Lovell, F.R.S.: "The Exploration of Outer Space" (Trueman Wood Lecture).

Royal Statistical Society (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.15 p.m—Mr. E. Strauss: "The Structure of the English Milk Industry".

Institute of Fuel (at the Institution of Civil Engineers, Great George Street, London, S.W.1), at 5.30 p.m.—Mr. E. H. Hubbard: "Comparison of Different Methods of Fluid-atomizing Oil Flames", and "The Effect on Flame Emissivity and Radiation of the Addition of Carbon Black to Liquid Fuels" (Performance Trial No. 8 of the Joint Committee of the International Flame Research Foundation).

Institution of Electrical Engineers, Electronics and Communications Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Prof. A. L. Cullen: "Applications of Microwaves".

University College (in the Anatomy Theatre, Gower Street, London, W.C.1), at 5.30 p.m—Prof. D. H. Wilkinson: "Language and the Elementary Particles".* (First of three Special University Lectures in Physics. Further lectures on February 25 and 26.)

Society of Chemical Industry, Food Group (joint meeting with the Microbiology Group, at 14 Belgrave Square, London, S.W.1), at 6.15 p.m.—Meeting on "New Methods of Factory Hygiene in the Food Industry". Mr. G. A. Thomas: "Chemical Sterilants in the Food Industry"; Mr. W. J. Blois-Johnson "Hygienic Considerations in Food Plant Design".

Pharmaceutical Society of Great Britain (at 17 Bloomsbury Square, London, W.C.1), at 7.30 p.m.—Dr. T. D. Whittet: "Pharmaceutical Fellows of the Royal Society".

Thursday, February 25

Central London Productivity Association, Working Relations Sub-Committee (at 21 Tothill Street, London, S.W.1), at 2.30 p.m.—Meeting on "The Factories' Act and the Work of the Factory Inspectorate".

Royal Society (at Burlington House, Piccadilly, London, W.1), at 4.30 p.m.—Prof. F. W. R. Brambell, F.R.S., Mr. W. A. Hemmings, Prof. C. L. Oakley, F.R.S., and Mr. R. R. Porter: "The Relative Transmission of the Fractions of Papain Hydrolysed Homologous γ-Globulin from the Uterine Cavity to the Fœtal Circulation in the Rabbit"; Prof. D. Lewis, F.R.S.: "Genetic Control of Specificity and Activity of the *S* Antigen in Plants".

Linnean Society of London (at Burlington House, Piccadilly, London, W.1), at 5 p.m.—Dr. G. Kuschel: "The Natural History of Juan Fernandez Islands"; Dr. M. W. Holdgate: "The Natural History in the South Chilean Islands".

Friday, February 26

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Prof. Alexander Kennedy: "The Scientific Lessons of Brainwashing".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Knoop Research Fellow (graduate in medicine or science), to work on cerebral enzymatic aspects and on the presence of chemical abnormalities in hereditary degenerative cerebral conditions—The Secretary, Institute of Neurology, The National Hospital, Queen Square, London, W.C.1 (February 25).

Lecturer (graduate of a British university, preferably with teaching experience) in Pure and Applied Mathematics in the Department of Building, Structural Engineering and Surveying—The Secretary, Hammersmith College of Art and Building, Lime Grove, London, W.12 (February 25).

Horticulturalist (male), to take charge of the Insecticide Department glasshouses—The Secretary, Rothamsted Experimental Station, Harpenden, Herts (February 29).

Keeper (graduate or holder of the Diploma of the Museums Association, a good knowledge of Western European archæology, and preferably an interest in the archæology of the Eastern Mediterranean) of Archæology—The Director, Leeds City Museum, Park Row, Leeds 1 (February 29)

Lecturer in the Department of Psychological Medicine—The Secretary, The University, Edinburgh (February 29).

Research Student in Metallurgy, for postgraduate work on liquid metals—The Registrar, King's College (University of London), Strand, London, W.C.2 (February 29).

Lecturer in Social Science, a Lecturer in Social Psychology or in Sociology, and 2 Leverhulme Research Fellows—The Registrar, University College, Singleton Park, Swansea (March 5).

Lecturer (with a good honours degree and teaching experience) in Chemistry at the Nigerian College of Arts, Science and Technology—The Council for Overseas Colleges, 12 Lincoln's Inn Fields, London, W.C.2 (March 5).

Research Fellow in Mathematics—The Secretary of University Court, The University, Glasgow (March 5).

Senior Lecturer/Lecturer in Fuel Technology in the School of Mining, Engineering and Applied Geology, University of New South Wales—Agent General for New South Wales, 56–57 Strand, London, W.C.2, and The Bursar, Post Office Box 1, University of New South Wales, Kensington, N.S.W., Australia, marking envelope "University Appointment" (March 5).

Assistant Professor (with high academic qualifications in physics or mathematics, and with experience in (*a*) experimental reactor physics, (*b*) the design of critical and subcritical experiments, and (*c*) safety precautions associated with (*a*) and (*b*), and with the use of highly radioactive sources) of Nuclear Science and Technology at the Royal Naval College, Greenwich—The Civil Service Commission, 17 North Audley Street, London, W.1, quoting S./5101/60 (March 10).

Lecturer (with a degree in veterinary science, together with postgraduate experience) in the Department of Veterinary Preventive Medicine at the University of Queensland, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, March 12).

Physiologist or Biochemist (honours graduate in physiology or biochemistry, with relevant postgraduate research experience) in the Cattle Research Laboratory, Division of Animal Genetics, Commonwealth Scientific and Industrial Research Organization, Rockhampton, Queensland, Australia, for the study of the adaptation of cattle to a tropical and subtropical climate—Chief Scientific Liaison Officer, Australian Scientific Liaison Office, Africa House, Kingsway, London, W.C.2, quoting Appointment No. 231/5 (March 12).

Lecturer in Physics (Cosmic Ray Group)—Prof. P. M. S Blackett, F.R.S., Physics Department, Imperial College of Science and Technology, Imperial Institute Road, London, S.W.7 (March 14).

Biochemists (2) (honours graduates in science (or equivalent qualifications), with suitable postgraduate research experience, preferably to Ph.D. level) in the Division of Biochemistry and General Nutrition, Commonwealth Scientific and Industrial Research Organization, Australia, for research into the intermediary metabolic processes of ruminant animals—Chief Scientific Liaison Officer, Australian Scientific Liaison Office, Africa House, Kingsway, London, W.C.2, quoting Appointment No. 250/92 (March 15)

LECTURER IN ZOOLOGY—The Secretary, The Queen's University, Belfast (March 15).

LECTURERS IN PURE MATHEMATICS—The Secretary, The Queen's University, Belfast (March 21).

SENIOR LECTURER (with special qualifications in hydraulics) IN CIVIL ENGINEERING at the University of Adelaide, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia, March 21).

CHAIR OF PSYCHOLOGY—The Registrar, The University, Leicester (March 26).

LECTURER IN PSYCHOLOGY—The Registrar, The University, Leicester (March 26).

CENTRAL ELECTRICITY GENERATING BOARD RESEARCH FELLOW (of British nationality. and by October 1960, taken the degree of Doctor of Philosophy, or have had equivalent research experience), for research in the physical sciences, including metallurgy, engineering or mathematics—The Master Elect, St. Catherine's College Planning Office, 42 Banbury Road, Oxford (March 31).

PROFESSOR OF EDUCATION at the University of Queensland, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (March 31).

SECRETARY (with senior administrative experience in or with Governments, preferably in departments connected with science, agriculture or forestry; also overseas experience or experience in inter-commonwealth relations, and preferably aged 50 to 55 with a scientific background or training, and fluency in some foreign languages) TO THE EXECUTIVE COUNCIL OF THE COMMONWEALTH AGRICULTURAL BUREAUX—The Secretary, Commonwealth Agricultural Bureaux, Farnham House, Farnham Royal, Bucks (April 15).

IMPERIAL CHEMICAL INDUSTRIES RESEARCH FELLOWS IN BIOCHEMISTRY, CHEMISTRY, CHEMOTHERAPY, ENGINEERING, METALLURGY, PHARMACOLOGY or PHYSICS—The Academic Registrar, University of London, Senate House, London, W.C.1 (April 30).

TURNER AND NEWALL RESEARCH FELLOW IN ENGINEERING, INORGANIC CHEMISTRY or PHYSICS—The Academic Registrar, University of London, Senate House, London, W.C.1 (April 30).

LECTURER (preferably with experience of terrestrial ecology) IN ANIMAL ECOLOGY—The Secretary, University College London, Gower Street, London, W.C.1 (May 1).

NATIONAL RESEARCH COUNCIL OF CANADA POSTDOCTORATE FELLOW IN APPLIED MATHEMATICS—G. R. Magee, Department of Pure and Applied Mathematics, University of Western Ontario, London, Ontario, Canada (May 1).

RESEARCH SCHOLARS (with a good honours degree or graduate member of the Royal Institute of Chemistry) IN THE DEPARTMENT OF CHEMISTRY, for research in higher dispersion infra-red spectroscopy for Ph.D. or M.Sc. degree—The Professor of Chemistry, The University, Reading (May 2).

HOLLERITH RESEARCH STUDENT (graduate of any university), to undertake research in automatic computing or in a subject allied thereto—The Secretary to the Academic Council, The Queen's University, Belfast (June 1).

AGRICULTURAL CHEMIST (male, with a good honours degree in agricultural chemistry, with some postgraduate experience, preferably in the tropics) in Sierra Leone, to advise the Director of Agriculture on specific problems and for investigational studies in the field, including the laying out of field trials and their statistical analysis and interpretation—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.63/15/04/T.

ASSISTANT or ASSOCIATE PROFESSOR OF MATHEMATICS—The Principal, Victoria College, Victoria, B.C., Canada.

BIOCHEMIST (Senior or Basic grade) to the Rochdale Group of Hospitals, to work under the Consultant Pathologist and to carry out biochemical work for the hospitals and clinics of the Group—The Group Secretary, Rochdale and District Hospital Management Committee, Central Offices, Birch Hill Hospital, Rochdale, Lancs.

CHEMIST (Ph.D.), to supervise rock and soil analytical laboratory, a CHEMIST (honours B.Sc.), to assist in the study of sulphur compounds in oils, a CHEMIST (B.Sc.) to assist in gas adsorption studies, a CHEMICAL ENGINEER (B.Sc. or M.Sc, with a bias towards mathematics) for basic combustion research, and a CHEMICAL ENGINEER (B.Sc. or M.Sc.) to work on pipe-lining research—The Secretary, Research Council of Alberta, 87th Avenue and 114th Street, Edmonton, Alberta, Canada.

EXPERIENCED PHOTOGRAPHER FOR THE PATHOLOGICAL AND BACTERIOLOGICAL RESEARCH LABORATORY—The Administrative Officer, Sir William Dunn School of Pathology, University of Oxford, South Parks Road, Oxford.

FISHERIES OFFICER (with a university degree in marine biology and suitable postgraduate experience) in the Bahamas, to advise the Department of Agriculture on fisheries conservation, introduce new fishing methods and enforce the fisheries laws—The Director of Recruitment, Colonial Office, London, S.W.1, quoting Ref. BCD. 63/27/04/T.

GEOPHYSICIST or GEOLOGICAL ENGINEER (Ph.D. or M.Sc.), to work on groundwater hydrology; and a GEOLOGIST or GEOLOGICAL ENGINEER (M Sc), to work on problems of applied groundwater geology and hydrology—The Secretary, Research Council of Alberta, 87th Avenue and 114th Street, Edmonton, Alberta, Canada.

MASTER TO TEACH PHYSICS to Open Scholarship level—The Headmaster, Bolton School, Bolton, Lancs.

NATIONAL RESEARCH COUNCIL AND OTHER POSTDOCTORAL FELLOWS in all branches of chemistry, and PRE-DOCTORAL TEACHING RESEARCH FELLOWS (well qualified graduate students), for research work for the M.Sc. and Ph.D. degrees—Prof. C. A. McDowell, Department of Chemistry, University of British Columbia, Vancouver, B.C., Canada.

PHYSICAL CHEMISTS (graduates with a good honours degree, preferably with some postgraduate experience or Ph.D. degree), to join a team working on fundamental studies on the physical properties of high polymers—The Director, Arthur D. Little Research Institute, Inveresk, Scotland.

PRINCIPAL FISHERIES RESEARCH OFFICER (with an honours degree in biology with not less than six years postgraduate research experience, preferably in fisheries science or marine biology) with the Federal Government of Nigeria, to plan and carry out research connected with the sea and inland fisheries of Nigeria, including studies in the field of population dynamics, and to supervise a small research group—The Appointments Secretary, Federal Public Service Commission, Nigeria House, 9 Northumberland Avenue, London, W.C.2, quoting H.1/1.

SCIENTIFIC OFFICER, CHEMIST (with a good honours degree in chemistry, and preferably with some experience in foods and oilseeds analysis, and a knowledge of fumigant and insecticide analysis) with the West African Stored Products Research Unit, Federation of Nigeria, to undertake research into problems associated with storage of oilseeds and other food crops with particular reference to insect infestation with the object of evolving methods for reducing losses—The Director of Recruitment, Colonial Office, London, S.W.1, quoting Ref. BCD.153/14/05/T.

SENIOR LECTURER IN ELECTRONICS, a LECTURER IN ELECTRONICS OR MEASUREMENTS, a LECTURER IN GENERAL ELECTRICAL ENGINEERING, and an ASSISTANT LECTURER IN ELECTRONICS—The Secretary, Northampton College of Advanced Technology, St. John Street, London, E.C.1.

REPORTS and other PUBLICATIONS

(*not included in the monthly Books Supplement*)

Great Britain and Ireland

General Register Office. The Registrar General's Quarterly Return for England and Wales. Births, Deaths and Marriages; Infectious Diseases; Weather; Population Estimates; quarter ended 30th September 1959. (No. 443, 3rd Quarter, 1959.) Pp. 20. (London H.M. Stationery Office, 1959.) 2*s*. net. [2212

National Union of Journalists Freelance Directory, 1959. Pp. i+80. (London: National Union of Journalists, 1959.) [2212

Agricultural Research Institute of Northern Ireland. Thirty-second Annual Report, 1958–59. Pp. 42. (Hillsborough, Co. Down: Agricultural Research Institute of Northern Ireland, 1959.) [2212

Department of Scientific and Industrial Research. Mechanical Engineering Research 1958: Report of the Mechanical Engineering Research Board with the Report of the Director of Mechanical Engineering Research. Pp. iii+67+4 plates. (London: H.M. Stationery Office, 1959.) 4*s*. 6*d*. net. [2212

Government of Northern Ireland: Ministry of Agriculture. Leaflet No. 103: Catch Crops. Pp. 2. (Belfast: Ministry of Agriculture, 1959.) [2212

Evaluation and Presentation of Spectro-Analytical Results. By Dr. A. B. Calder. Pp. 47. (London: Hilger and Watts, Ltd., 1959.) 5*s*. net. [2212

Other Countries

National Science Foundation. American Science Manpower. Employment and other Characteristics, 1954–55. (Based on the National Register of Scientific and Technical Personnel, 1954–55.) Pp. vi+48. (NSF 59–39). Scientific Manpower 1958: Papers of the Symposium on Demographic and Sociological Aspects of Scientific Manpower. Pp. v+87. (NSF 59–37). (Washington, D.C.: National Science Foundation, 1959.) [2212

Union of South Africa: Department of Commerce and Industries. Report No. 37: The South African Pilchard (*Sardinops ocellata*) and Maasbanker (*Trachurus trachurus*). Hydrographical Environment and the Commercial Catches, 1950–57. By M. E. L. Buys. Pp. 176. (Reprint from *Commerce and Industry*). (Pretoria: Government Printer, 1959.) [2212

Report on the Administration of the Meteorological Department of the Government of India in 1953–54. Pp. 76+3 plates. (Simla. Government of India Press, 1956.) [2212

Indian Forest Bulletin No. 215 (New Series): Entomology. Prophylactic Efficacy of Various Insecticides in the Protection of Freshly Felled and Converted Timbers (Planks) Against Insect Borers. By M. L. Roonwal, P. N. Chatterjee and R. S. Thapa. Pp. ii+45. (Delhi Manager of Publications, 1959.) Rs. 4.12; 6*s*. 6*d*. [2212

India. Council of Scientific and Industrial Research: Central Fuel Research Institute. Report of Director for the year 1957–58. Pp. iv+115. (Jealgora Dhanbad, Bihar: Central Fuel Research Institute, 1958.) [2212

Indian Forest Bulletin No. 171 (2) (New Series): Entomology. A List of Insect Pests of Forest Plants in India and the Adjacent Countries (Arranged Alphabetically According to the Plant Genera and Species, for the Use of Forest Officers). Part 3: List of Insect Pests of Plant Genera "A" (Appendix only), "B" (*Baccaurea* to *Buxus*) and "C" (in part) (*Cadaba* to *Citrus*). By G. D. Bhasin, M. L. Roonwal and Balwant Singh. Pp. i+126. (Delhi: Manager of Publications, 1959). Rs. 4.50, 7*s*. [2212

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UNITED NATIONS COLLABORATION IN THE SOCIOLOGICAL SCIENCES

SOME ten years ago Mr. L. V. Berkner, as special consultant to the Department of State, Washington, surveyed the Department's responsibilities in international science. The subsequent report, issued in May 1950 and entitled "Science and Foreign Relations", made numerous positive recommendations to promote the utilization of science in shaping national policy, including the establishment of a science office in the Department of State, the appointment of scientific attachés and measures to encourage the interchange of scientific and technical personnel and the flow of scientific and technical information. The report was discussed at the time in these columns (*Nature*, 166, 81; 1950) and on subsequent occasions (for example, 170, 1033; 1952), and the importance of its recommendations has become increasingly apparent with the growth of technical assistance programmes and of other measures to assist the under-developed countries.

In January 1958, the United States Committee on Foreign Relations decided to undertake a review of conditions and trends in the world, and of the relevant policies and programmes of the United States, prompted partly by the impact which Soviet scientific achievements might have upon the relations of the United States with the rest of the world. The inquiry then instituted developed later into a series of some fifteen studies of various aspects of the problem designed to provide a simple forthright statement of the basic aims of the foreign policy of the United States, which would reflect the motives and aspirations of its people, identify the forces which may frustrate or promote those aims, suggest means of controlling those forms and examine their impact upon the conduct of American foreign policy and the adequacy of the organization for determining such policy. The first of these studies, dealing with "Economic Problems and their Impact", was issued in August 1959, but the second, entitled "Possible Non-Military Scientific Developments and their Potential Impact on Foreign Policy Problems of the United States"*, is of as outstanding interest to the scientific and technical world generally as it is to those concerned with assistance to the under-developed areas of the world (to which one section of the report is devoted), and more especially with technical assistance, and it merits close attention as fully as the original Berkner Report.

This study was made by the Stanford Research Institute for the Committee on Foreign Relations, and it explores, besides prospective scientific developments of the next decade and their potential impact on foreign policy, the possibility of making more positive use of science and technology. It does not except for brief discussion, deal with possible scientific contributions to problems of disarmament or examine developments in military technology; nor does it deal further with matters such as scientific attachés, the promotion of scientific exchanges and the support of scientific activities overseas, with which the Berkner Report itself was concerned. Broadly, the report points out that while scientific developments may help to solve some problems of foreign policy, unless deliberate measures of policy are taken, the progress of science and technology will do more to create or intensify than to solve such problems. In the absence of such measures, the combination of pressures may easily lead to their relief by the traditional methods of war, famine and disease. The great danger is complacency, for under present allocations of effort to research and the diffusion of results, the urgency of the problems is likely to increase faster than the means of solving them.

The national interest of the United States thus requires a more conscious direction of scientific activity in ways which promise to assist in attaining the country's international objectives. In other words, security and welfare themselves call for a re-appraisal of present allocations of scientific and technical effort so as to increase that directed toward meeting the non-military challenges of foreign policy. The report suggests that the Committee on Foreign Relations could take steps to stimulate additional research and development directed specifically at important problems in that field, and indicates particular types of problem which call for such attention. It also suggests that steps could be taken to double or triple the amount of fundamental scientific research in fields where progress in scientific understanding is particularly important to foreign policy, and that the Committee should encourage the international proclivities of science and scientists to build bridges of understanding. Concrete suggestions include more international scientific conferences and interchange of personnel, international research programmes, an international centre for translation and scientific information, a world-wide attack on the problems of economic and social development in an 'International Development Year', and a lead in establishing a United Nations university. Foreign policy planning of the broadest kind, utilizing the best scientific assistance, will be a critical requirement in the years ahead, and this should include continual review of prospective scientific developments and their significance for international relations.

In this report the relations between scientific developments and foreign policy are discussed in the first chapter, including both the way foreign policy has been affected in the past, the nature of the connecting links and the possibilities both of anticipating and shaping scientific developments. The second section, dealing with future problems of foreign policy in the light of scientific developments, which in successive chapters covers science and national sovereignty; population, food and science; science and the under-developed countries; energy, materials and trade; and science and international communications; it approaches these problems primarily but not exclusively from the point of view of the United States. Nevertheless, like the section on future policy, where the positive use of science to achieve international goals is more particularly considered, it contains much that is fully relevant to those concerned with science and with foreign policy in other countries, and the whole report deserves the closest attention not only of the Secretary of State for Foreign Affairs in the United States but also of the Minister for Science and the Advisory Council for Scientific Policy in Great Britain. The picture displayed of the immense array of complex interlocked problems calling for urgent action applies fully as much to Britain, and it may be even more imperative for both the security and the welfare of Britain that the right priorities should be determined and our limited resources applied in accordance with them.

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* United States (86th Congress, 1st Session). United States Foreign Policy. Possible Nonmilitary Scientific Developments and their Potential Impact on Foreign Policy Problems of the United States. A Study prepared at the request of the Committee on Foreign Relations, United States Senate, by Stanford Research Institute. Pp. ix+100. (Washington, D.C.: Government Printing Office, 1959.)

This is especially evident in the chapter of the report which considers the positive use of science to achieve international goals. The evidence presented there of the accelerating rate of discovery and invention applies generally. All over the world the number of scientists and technologists is rapidly increasing, and also expenditure on research and development. The increasing efficiency of diffusion of new ideas and techniques, and the more powerful tools of investigation available to scientists and technologists, act in the same way as does the expansion of organized team-work and the more favourable social attitudes, while to-day international competition and emulation tend to spur scientific and technical development and could easily do so in ways wasteful of the world's resources. Nevertheless, there are fields in which scientific effort is still undesirably low, and the report remarks on the effort specifically directed towards the peaceful achievement of objectives of foreign policy and the basic problems of human relations and social organization, including international aspects of such problems.

The report, in discussing next the positive use of science, makes some specific suggestions regarding non-military aspects of problems of foreign policy in which insufficient attention has been given to the possibility of attack by adequate fundamental research in the relevant sciences. It suggests, for example, that a responsibility should be established within the Government for continuing review of prospective scientific developments and their impact on foreign policy. In Britain, this would now appear to rest unequivocably with the Minister for Science and the Advisory Council for Scientific Policy. More specifically, the report quotes problems in oceanography, the polar regions, the atmosphere and space research, but stresses the need to include also studies of a legal, political and administrative as well as physical and biological character. Again, it is asked whether sufficient positive use of science is made in developing and adapting promising methods of improving food production, making the most effective use of American surpluses and exploring the possibilities of population control. Next, there are problems of planning for new world relations in production and trade, such as assessing the new competition and market opportunities likely to result from the spread of modern technology to the formerly under-developed countries, or the possibility of making long-term analyses of world supplies of raw materials and energy, and of world trade patterns. Problems of communication with other nations provide another set of problems in social science, and there are others of international political organization, law and administration where further research on political systems and legal and administrative systems might disclose the best ways of strengthening the United Nations and advancing towards a world rule of law.

In two particular sections, the report deals at greater length with the possibilities of action and the range of problems presented. First, in the matter of arms control and disarmament, besides the technical problems of control of armament and the devising of systems of inspection to prevent violation calling for constant research, the economic repercussions and adjustments should be explored, so that the individual nations are ready to deal with the consequences should a substantial measure of disarmament become possible. The report emphasizes the need for a continuing study, adequately financed, to deal with such technical problems as observation techniques, including radar, aerial photography, ground observers, seismic means for monitoring nuclear explosions, problems involving communication and data-processing systems.

Secondly, the development of under-developed countries presents a wide range of problems in which specific research is likely to assist materially. The report mentions simpler and cheap power sources for villages and towns, including electrical generators from local resources, gas from agricultural wastes, etc., wind-power, solar energy, storage of energy and small nuclear power stations. Improved but simple and cheap housing materials and constructional methods offer other possibilities, as does water supply for irrigation and industrial use, for example, by cheap desalting processes. Simple, cheap methods of refrigeration for foods and cooling of houses; environmental sanitation; the control of special diseases, such as hepatitis, filariasis, brucellosis, trachoma, dysenteries; food preservation; the improvement of food crops and animals; and the control of the growth of population—all these present physical science with urgent research problems.

Attention is also directed to the range of problems in sociological and social research presented by the improvement in techniques of planning development programmes, by development of simple, cheap methods of mass education, the diversity of methods of introducing agricultural and rural improvements, communicating effectively with people, and inducing them to adopt better practices. Methods of introducing modern industrial techniques and meeting problems of industrialization, and government methods compatible with democratic ideals but more suitable to the special circumstances of the under-developed countries, offer another fruitful field for such research, and much emphasis is laid not only on the need for fundamental research in the physical, as well as in the pyschological and social sciences, but also for better theories. The major need, it is asserted, is for better interpretation of the facts, better understanding of their meaning in terms of our aims, ideals and plans, and for better founded notions about the probable effects of one course of action or another.

It is not suggested in the report that simple solutions to these problems are likely to be found, but there is sound sense in the argument that it is at least possible that imaginative, carefully designed basic research could eventually throw light on the soundness of even such a difficult problem as the foundations of the psychological assumptions of the theory of deterrence on which much of the Western policy for preventing nuclear attack is based. Excluding the military aspects, the problems of foreign policy are overwhelmingly human ones, and the psychological and social sciences, to which the makers of foreign policy should be able to, and, to a significant extent do, look for insight and help, are the least well developed and least well supported of the major scientific groups. Without expecting quick and easy solutions to problems of human relations, there is ample basis for believing that more effort in this field could lay the foundations for much better understanding of the nature of some of the problems that arise in foreign policy and considerably help those who make it. The president of Stanford Research Institute, Mr. F. Finlay Carter, unhesitatingly affirms his belief that research can make many contributions to the better understanding of the invisible but very real forces affecting human behaviour, human relations and human institutions.

In view of the establishment last year in Britain of the Overseas Research Council, it is interesting to note that the report suggests that the United States Government should be provided at some appropriate point with an office of research and development concerned with problems of under-developed areas, and that such an agency could initiate a systematic programme for identifying problems where scientific and technological discovery could assist to accelerate development and that it could then stimulate research on these problems, either in the United States or abroad. Even in the physical sciences, the problems of this type are in some way or other neglected in the post-war world; and it is obvious that their solution must involve some redistribution of resources.

That is one sound reason for advocating that the attack upon them, where possible, should be made by international agencies; and not the least important section of this highly suggestive report is that where, in conclusion, it discusses the possibility of encouraging the tendencies of science and scientists to work across national frontiers to build bridges of understanding between East and West—between the under-developed and the industrially advanced nations. The value of scientific conferences and the interchange of scientists and scholars at all levels is recognized in the first place, and some stress is also laid on the contribution which advances in mechanized translation and information facilities could make both by improving communication between scientists and reducing the language problems of mankind. Moreover, it has already been widely recognized that some of the fields of research to which the report directs attention can be prosecuted to advantage by international agencies or by the co-operation of research institutions in several countries. Examples are to be seen in oceanography, polar and space research, and the atmosphere; and the report of the International Council of Scientific Unions indicates not merely that a start has already been made, but also something of the possibilities, given substantial financial backing and goodwill. Arctic and deep-sea research are instanced as further possibilities which merit examination, and also the establishment of a United Nations weather and climate agency.

The proposal regarding the under-developed countries already noted deserves special attention as indicating a novel approach to a problem of growing urgency and importance. The International Development Year suggested would initiate, much on the lines of the International Geophysical Year, fundamental new studies on an international basis on the key development problems of under-developed countries, and would stimulate the fundamental research in these aspects of the physical, biological, psychological and social problems from which contributions to the solution of these problems might be expected. The United Nations university scheme, which, it is suggested, the United States might also take the lead in establishing, would possibly include a World Institute of Social Technology, for advanced research and teaching in the practical application of psychological and social science to problems of human relations and social organization, especially where they bear on these problems; but the major purpose of the university scheme would be the advancement of science and the humanities, and thereby the discovery and development of the unifying elements in the human values widely held by the different groups and cultural traditions of mankind. It is suggested that the system might be financed by an annual contribution of 0·1 per cent of the annual expenditure of each member of the United Nations on military defence, or an agreed fraction of the index of the total national product.

No reader of the report is left in any doubt as to the danger of failing to give much greater attention to the significance of scientific and technological advance for national policy, including foreign policy, or as to the improbability that an adequate attack can be made on many of the problems now confronting us except on an international basis. The facts that the creation of a Minister for Science and the existence of an Advisory Council for Scientific Policy in Britain, together with the clear recognition that Britain's contribution to space research should be made on an international basis, place this country in an advantageous position to co-operate in this way, is no ground for complacency. The suggestions made in this report and their implications deserve to be closely considered by the Minister for Science, the Advisory Council for Scientific Policy and the Overseas Research Council at least, with a view both of increasing our present effort and ensuring that it is concerted as effectively as possible in co-operation with that of other nations.

POST-MORTEM ON GROUNDNUTS?

Agriculture and Ecology in Africa

A Study of Actual and Potential Development South of the Sahara. By Prof. John Phillips. Pp. 424. (London: Faber and Faber, Ltd., 1959.) 63*s*. net.

POLITICAL advances in tropical Africa can only be successful if economic progress is accelerated. Industrial developments are important, but for many years agriculture in one form or another will be the major industry of all the countries in Africa south of the Sahara. Today agricultural progress is slow, and where crops have brought prosperity, as cocoa has to Ghana, this is because the price has risen disproportionately to the rise in other world prices, and not because the yield has risen substantially. Unless methods of agriculture in many countries are greatly improved, as the population increases even the present pitifully low standards of living will be difficult to maintain.

Prof. Phillips's book is about this problem. He has been himself concerned with ecological and agricultural works in many parts of Africa for nearly forty years. He was the botanical ecologist in Swynnerton's team of tsetse fly investigators in Tanganyika. He was professor of botany at the University of the Witwatersrand. He was one of the senior scientists concerned with the ill-fated East African groundnut scheme. Since 1952 he has been professor of agriculture in the University College in Ghana. Few men can have seen so much of African agriculture, and have been made so aware of its problems.

In this book an immense amount of information about the climate, soil, crops and people of Africa is brought together. It must have been a tremendous task for the author to make this compilation, and his book will at least serve as an invaluable work of reference to those unable to go to the original sources. I do not think that it will succeed so well in its avowed purpose "to awaken those concerned with Africa to the necessity of adjusting both traditional and modern agriculture to the ecological, sociological and psychological settings of the Trans-Sahara".

The first disadvantage of this volume is that it is so difficult to read. The (very valuable) data are set out at length, usually undigested, and I found I could only assimilate those in which I had some particular and personal interest. Between the data the reader is treated to infuriating examples of flowery prose in Prof. Phillips's own particular style. In one paragraph three consecutive sentences start: "Sketchy is the information . . .". "Essential is the need . . .". "Unnecessary is a review . . .". We are, however, left with a true picture of Africa, vast, varied, and (in the main) unproductive; but this could have been much more easily achieved and with less strain on the reader.

What should have been the most valuable chapter compares three large-scale development schemes. These are the Sudan Gezira Scheme, the East African Groundnut Scheme, and the proposed Volta River Project in Ghana. We are shown how fundamentally different the conditions for the successful Gezira Scheme and the groundnut fiasco were, but instead of a straightforward account of each we are treated to a point-by-point comparison of features which are not always strictly comparable. We can in the end only conclude that in the Gezira conditions were favourable, and in Tanganyika everything was done in too great a hurry.

It is difficult to imagine how this book can have any important and practical results. We are told that agriculture must be ecological, which simply means that we must learn to grow the correct crops by the best methods in the best places. We are not told how to do this, except that we must not be in too much of a hurry, and that research must be properly co-ordinated.

In my opinion the trouble with African agriculture is not that information is not properly co-ordinated, but that the basic facts are simply not known. So little fundamental agricultural research has been done. Even in Britain we spend more in subsidizing our poultry industry for a month than we spend on agricultural research in a year; in Africa, with its immense areas and problems, we spend much less. Even attempts at developments like the Groundnut Scheme, into which millions of pounds were poured, seem less wasteful when it is realized that for every pound lost in this and in all agricultural schemes attempted in Africa since the War, much more than a hundred pounds has been spent in subsidizing British agriculture at home. More money will have to be spent on research in Africa. We need to have more and better research workers in the laboratories and in the field; I believe they will use the limited resources better than those who fritter away so much time and money attending international conferences and endless meetings of co-ordinating committees which lack the basic facts to co-ordinate. If the newly independent territories in Africa can learn this lesson, and develop an environment favourable to working scientists (expatriate as well as local), there is a reasonable chance that they may learn to take proper advantage of the ecological conditions of their countries in agriculture as in other fields of development.

KENNETH MELLANBY

CHANGING COASTLINES

Beaches and Coasts

By Dr. C. A. M. King. Pp. xii+403. (London: Edward Arnold (Publishers), Ltd., 1959.) 65*s*. net.

IT is now forty years since the publication of D. W. Johnson's pioneer and now classic work "Shore Processes and Shoreline Development". In the meantime much work has been carried out, both theoretica and practical, on the processes at work along the coasts and the evolution of different types of shoreline Problems of coast erosion and, equally, of coasta accretion have long been matters of much concern to civil engineers who, in the absence of precise investigations, were compelled to use empirical methods. Work in recent decades has been full of surprises. Though the widespread occurrence of steep-sided offshore canyons around the coasts is one of the surprises, this lies beyond the scope of the present work, where a beach is defined as an accumulation of loose material around the limit of wave action—extending through a zone divided into backshore, foreshore and offshore, from the extreme upper limit of wave action to the depth at which waves approaching from deep water first cause appreciable movement of the bottom materials. This scholarly, well-documented treatise first defines the four main factors on which the character of a beach depends, namely beach material, waves, tides, winds. It then considers briefly the three parallel methods of research—theoretical, experimental (especially wave-tanks such as that long in use in the Department of Geography at Cambridge) and field observations. A fourth approach, the historical, is mentioned, but sufficiently detailed information is rarely available. The main topics are then taken up in the order of waves, movement of material, beach profiles, effect of wind, classification of beaches and coasts, constructive and destructive wave action, and, finally, the marine cycle. Examples are drawn from world-wide sources with a particularly thorough examination of all the work around Britain.

Every point is substantiated by appropriate references, and nowhere is the reader left with the impression that all is adequately known and understood. Instead, the memory of the disastrous coastal floods of 1953 is still fresh in England; what may prove to be the tragic mistake of closing to scientific research the great natural laboratory of Dungeness is within the last few months. There is no doubt that protective works in one area have unexpected results in another; if half a million acres of tidal flat are awaiting 'reclamation', as some would claim, it is vital to know what will be the consequences elsewhere. The field of study covered in this book is part of the important new field of applied geomorphology. It is perhaps inevitable, if regrettable, that a new 'jargon' should arise that one needs to study the dilatancy and thixotropy of sediments. But perhaps the deliberate creation of new words is safer than the attempt to define by size granules, pebbles, cobbles and boulders, or to insist on new exact meanings for berm and runnel.

L. Dudley Stamp

PRECIS OF PROGRESS IN BIOCHEMISTRY

Progress in Biochemistry

A Report on Biochemical Problems and on Biochemical Research since 1949. By Prof. Felix Haurowitz. Pp. xii+357. (Basel: S. Karger; New York: Interscience Publishers, Inc.; London: Interscience Publishers, Ltd., 1959.) 60*s*.; 8.50 dollars.

THE fifth in Prof. Haurowitz's series of reports on progress in biochemistry deals with the advances in this field since 1949, and we gather from the author's foreword that he was hesitant to prepare it. He comments that "limits of time and human endurance make it more and more difficult to grasp, let alone to master, the entire field of biochemistry". This *cri de cœur* will surely find an echo in many laboratories and lecture rooms, and for this reason alone, quite apart from the book's intrinsic merits, it is likely that Prof. Haurowitz's courage and virtuosity in grappling with the smothering excess of the printed word will earn the reader's gratitude.

But his claim to our thanks is more firmly founded for there can be no doubt that, within his terms of reference, he has written a most useful book, which gives fluent accounts of the progress made in all those aspects of the subject normally dealt with in the standard text-books. His method has its own particular merits: thus, except where a brief reminder may help the correlation of recent advances with established work, it is free from the necessity of giving the repetitious background to knowledge which necessarily swells each general text-book in turn. On the other hand, because it reports the work of a decade it is frequently able to focus topics which an annual review sees only as the indistinct blur of work-in-progress; moreover, it is spared the uncritically comprehensive list of references which can so effectively throttle the text of more definitive reviews. In ten years a measure of sifting becomes possible, and the references retained and commented upon here seem to have been chosen with excellent discrimination.

Taken on this level, therefore, as a series of timely supplements to the general text-books, these volumes may be judged to have introduced successfully a new style of reporting into biochemistry. Necessarily they risk the defects of any anthology; there may be omissions and faults of emphasis. But it is certain that most readers will find new and stimulating expositions in the pages of this latest volume, and even be led to an appreciation of new aspects of the subject which had lain unregarded by them in the more specialist literature.

F. H. Malpress

APPLIED MICROBIOLOGY

Advances in Applied Microbiology

Vol. 1. Edited by Wayne W. Umbreit. Pp. xi+304. (New York: Academic Press, Inc.; London: Academic Press, Inc. (London), Ltd., 1959.) 9.50 dollars.

THIS volume is presented as the first of a projected new series of reviews on microbiological topics of interest to the practical microbiologist. Applied microbiology is, in its widest sense, a subject that covers or overlaps considerable areas of present-day scientific research. In recent years the synthetic and degradative powers of micro-organisms have been increasingly harnessed in the laboratory and factory, as microbial synthesis has often proved the only convenient method of preparing a number of compounds important in pharmacy or nutrition.

Three chapters are on general problems of large-scale fermentation: P. Gerhardt and M. C. Bartlett classify the types of continuous fermentation and consider their theoretical basis and practical applications, M. Herold and J. Nečásek discuss the value of the use of antimicrobial agents to control contaminating micro-organisms that interfere with industrial fermentation processes, and R. F. Robinson and R. S. Davidson give an interesting account of the problems attending the large-scale production of edible fungi and urge greater consideration of the use of the higher fungi to convert carbohydrate-containing wastes into foods rich in fat and protein. In the three articles concerned with the use of micro-organisms as synthetic tools D. Perlmann gives a comprehensive and fully documented account of the biosynthesis of vitamin B_{12} and its analogues, A. L. Demain contributes a critical discussion of the biosynthetic pathway leading to penicillin, with particular reference to the synthesis of the β-lactam-thiazolidine ring nucleus, and S. Kinoshita introduces the new and rapidly developing field, largely pioneered in Japan, of amino-acid production by fermentation processes.

Articles dealing with the control of micro-organisms include a discussion of the antimicrobial activity of phenols by E. O. Bennett, a brief review of the use and mode of action of antibiotics in the control of plant diseases by D. Pramer, and a general outline of some of the problems associated with the application of radiation preservation to food and pharmaceutical products by W. D. Bellamy. A. W. Phillips and J. E. Smith trace the development of the methods used for the rearing of germ-free animals, list their characteristics and consider some of the many uses to which such animals have been, or can be, put. In the remaining chapter, S. R. Dutky contributes a valuable discussion of the application of specific micro-organisms in the control of insects, a procedure that eliminates many of the hazards to plants and animals presented by the use of insecticides.

The eleven chapters in this book deal with a representative selection of topics, and the diversity of the subjects is no doubt responsible for some of the differences apparent in the level of treatment in individual articles. The book is well presented, adequately indexed and should surely be welcomed as the first of a useful series.

J. W. G. PORTER

TRAINING APPRENTICES

Apprenticeship

An Enquiry into Its Adequacy under Modern Conditions. By Kate Liepmann. (International Library of Sociology and Social Reconstruction.) Pp. x+204. (London: Routledge and Kegan Paul; New York: The Humanities Press, Ltd., 1960.) 23s. net.

FOR two years a small group of investigators from the University of Bristol made an inquiry into systems of apprenticeships in firms in the Bristol area and in an agricultural area in North Somerset. Information was obtained from both the managerial and trade union sides of industry, as well as from technical colleges and relevant Government departments. Dr. Kate Liepmann was directly responsible for the study and here sets down details of the way the inquiry was made and a record of the findings.

The industries investigated—engineering, printing and building—were selected because of their local importance and because of significant differences in apprenticeship conditions, degrees of mechanization, 'de-skilling' and attractiveness to boys. An industry without apprenticeship, boot and shoe manufacture, was included for comparison. Most of the firms approached readily provided the information requested by the investigators. One large printing firm refused any information after a preliminary interview. Another big printing firm and the area's biggest engineering firm allowed the research workers access to certain senior executives but to no other employees. Information from these was obtained in other ways, but the omissions detracted from the value of the survey.

There is no reason to reject Dr. Liepmann's belief that the observations made during her Bristol survey would not be applicable to Britain as a whole. Its value is that this is an investigation carried out by professional research workers with no axe to grind, holding allegiance neither to employers, trade unions, technical college principals, nor Government agencies, and concerned only to record their findings quite impartially.

From the nation's point of view the scene described is depressing to contemplate. Instead of a means to build up the quality of human beings and to increase the country's resources of skilled craftsmen, the apprenticeship system is used by both sides of industry to further their respective sectional interest. "All the modifications made in adjustment to technical and other developments have been made in such a manner as to preserve the power of employers and trade unions to bend apprenticeship to their own ends".

The criteria for judging the workings of the apprenticeship system should be examined to see whether it supplies sufficient workers with the requisite skills, is efficient and economical to operate, and its effect on productivity. Dr. Liepmann's investigations show that industry is not training nearly enough apprentices with a wide degree of skill which will enable them to adapt themselves to changing technological needs. Nearly every firm looks after its particular interest and many of them rely on 'poaching' craftsmen from other firms to supply their needs. She rightly concludes that it is not in the best interests of the community that the number of apprentices should be determined by industry itself.

As a contribution to productivity the apprenticeship system leaves much to be desired. Although, as part of their training, apprentices make some contribution to productivity, the apprenticeship scheme leads to a widespread system of restrictive practices; rigid demarcation between trades impedes good manpower utilization, thereby lowering efficiency and increasing the cost of production: and, unfortunately, the demarcation which was formerly exclusive to apprenticed craftsmen is now being extended to a much wider class of workers, with further damage to productivity.

Despite the hope that some developments in the trade union movement may lead to the emergence of an attitude more in line with modern conditions, Dr. Liepmann concludes her inquiry with the firm belief that the State should take the prime responsibility for training for industry, both in its quantitative and qualitative aspects.

This is the second of two recent studies of the workings of the apprenticeship system by academic workers. Their observations are almost identical and should cause wide-spread concern. If the Ministers of Labour and of Education could be persuaded to read this or Lady Williams's book, they would see that the exhortations to industry of their predecessors have been so much wasted breath. Without a change in policy for the training of young workers, within twenty years Britain may face grave shortages of skilled and adaptable craftsmen.

Out of Doors

Edited by Winwood Reade. Pp. 160. (London: Rathbone Books, Ltd., 1959.) 12*s.* 6*d.* net.

"OUT OF DOORS" is a book written specially for children with the purpose of catching their interest and encouraging their observations of wild life. It cannot fail to do either. By dividing the book into twelve chapters, one for each month of the year, it provides a perpetual stimulus.

Under each month there are four sections. The first deals with things that can be seen out of doors. A large number of general facts are given here, as well as the specific observations for the particular month. The easy style and abundant illustrations of this first section set the tone for the rest. Secondly, there is an excellent article on one particular topic, written by a well-known naturalist. The third section of each month describes, very simply, something practical that can be made, done or collected. The last section is always two picture puzzles. These may be difficult for children; but even if they do not actually know the answers they will enjoy guessing, and learn something by looking up the correct answers in the back of the book.

The book combines all the qualities that enable a child to learn enjoyably. Not only is it highly informative, and infectious in its enthusiasm for the subject; it also appeals to a child's love of practical work, and encourages him to use his eyes. These qualities make "Out of Doors" suitable for even the young child. At the same time, many adults will be more than pleased to widen their knowledge of birds, trees, flowers, insects, animals, etc., in such a straightforward manner.

There are no coloured plates, which one misses, perhaps, in a book of this kind, and which always catch the eye, but this is more than made up for by the numerous excellent black-and-white illustrations. B. J. G. MAITLAND

The Waterfowl of the World

By Jean Delacour. Vol. 3: Eiders, Pochards, Perching Ducks, Scoters, Golden-eyes and Mergansers, Stiff-tailed Ducks. Pp. 270+20 plates. (London: Country Life, Ltd., 1959.) 126*s.* net.

WITH this third volume Mr. Delacour completes his account of the ducks, geese and swans of the world; we thus now have a comprehensive modern work on the Anatidae by a recognized authority, with Mr. Peter Scott's colour figures of the birds in their various plumage phases and useful distribution maps. The information given for each species or sub-species covers description, distribution, general habits and record in captivity—this last of special interest to the aviculturist.

Vol. 3 begins with the eiders, including the familiar British species. It proceeds to the pochards, narrow-billed and broad-billed respectively. Next come the perching ducks (or tree ducks), including the Carolina wood-duck so familiar in captivity and the beautiful mandarin duck which has lately become feral in England; the rest are tropical, including the so-called pygmy geese, the unique comb duck, the spur-winged goose, and the muscovy duck—the last a South American species which has been domesticated and is often crossed with the domestic mallard. Then we come to a group which includes such largely marine diving ducks as the scoters and golden-eyes and also the saw-billed ducks or mergansers. Finally there are the stiff-tailed ducks, including the strange musk duck of Australia and the aberrant black-headed duck of South America, which has the cuckoo-like habit of foisting its parental duties on other species.

Looking at this fine work as a whole—all three volumes—one is impressed by the great range of interest to be found within a group so relatively uniform that most of its members are included in a single sub-family. It is gratifying to have the information so competently and handsomely brought together. LANDSBOROUGH THOMSON

Uganda in Black and White

By Hugh B. Cott. Pp. xxvi+232 (109 figures). (London: Macmillan and Co., Ltd., 1959.) 30*s.* net

AN unusual and delightful volume which seeks to fascinate the reader with the beauties of Uganda and its inhabitants by means of no less than 109 full-page drawings with brief explanations on the opposite pages. There is a short foreword by Sir Andrew Cohen, a former governor of the Territory, and a not much longer introduction by the author, and then we come to the pictures. Dr. Cott is interested in crocodiles, and the main reason for his sojourn in Uganda was to study the habitat of these unpleasant beasts. But he is also an artist with a keen eye for the country and the people. Much more can be done with drawings than is possible with photographs. The latter are doubtless factually correct, but there is something that drawings can give which the unselective photograph cannot. Dr. Cott is interested in the old Uganda, not in its Westernized modern developments. He does not pretend to have visited every single part of it, but he does give us pictures from many regions. There are pampas swamps and country scenes, dhows on Lake Victoria, a Muganda lady, and, of course, some crocodiles. A lovely picture of the Ripon Falls before they vanished when the new dam was built can be taken in conjunction with another two of the Murchison Falls. Then there are giraffe browsing and a serval cat, not to mention elephants, rhinoceros, and and an Ankole cow. Some of the characteristic carvings in hard wood of various animals, etc., are also illustrated, as well as the people themselves. Altogether a delightful volume which it is a pleasure to possess. M. C. BURKITT

A Geography of Ghana

By E. A. Boateng. Pp. xvi+205+24 plates. (Cambridge: At the University Press, 1959.) 21*s.* net.

ALTHOUGH primarily intended for school examination candidates and first-year university students, this book deserves a wider public. Written by a Ghanian, who is now senior lecturer in the University College of his own country, for his own people, it reaches a high level in selection and presentation of material. An enclave surrounded by French-speaking territory, there has been some tendency for Ghana to be surveyed and studied in isolation, and it is useful to have summaries of local work in geology (with explanation of the special terminology), vegetation, soil and agriculture. A third of the book is devoted to a systematic treatment of the contrasted geographical regions. An excellent series of 41 photographs and 46 text figures and maps, all carefully prepared, completes a book of which Ghana may well be proud. L. DUDLEY STAMP

THE CERN PROTON SYNCHROTRON

By J. B. ADAMS

IN a recent article in *Nature*[1], in which a short review of the work of the laboratories of the European Organization for Nuclear Research (CERN) at Meyrin near Geneva, Switzerland, was given, it was mentioned that a 25-GeV. alternating gradient proton synchrotron was in an advanced stage of construction (Fig. 1). This accelerator has now been put into operation. Towards the end of November 1959, protons were accelerated up to 24 GeV. kinetic energy, and a few weeks later, after adjustments had been made to the shape of the magnetic field at field values above 12,000 gauss by means of pole-face windings, the maximum energy was increased to 28 GeV. The intensity of the accelerated beam of protons was measured as 10^{10} protons per pulse and there was no noticeable loss of particles during the whole acceleration period up to the maximum energy. The CERN proton synchrotron has thus fulfilled the expectations of its designers and the hopes of the twelve European countries that have supported the work for the past six years. It seems appropriate on this occasion to give a short account of this project together with a brief description of some of the design problems and preliminary measurements of the operating characteristics of this accelerator.

In June of 1952, at a conference in Copenhagen, the Interim Council of CERN decided to start an engineering design study of a 10-GeV. proton synchrotron that was to be the principal high-energy accelerator for nuclear physics research in Europe. The cost of this machine was considered to be so large in relation to other expenditures in physics research that no one European country felt justified in financing it alone. Hence several countries had combined their resources in order that physicists should have available in Europe the highest-energy accelerator that was thought feasible at that time. By July 1952 a design group of three people was set up with Dr. O. Dahl in charge, and in August this group, during a visit to Brookhaven on Long Island to see the cosmotron at first hand, learnt of a new idea for making cheaper and higher-energy machines. This idea was considered so attractive that in October 1952 the CERN Council was persuaded to drop the engineering study of a 10-GeV. proton synchrotron and instead to set up a theoretical study of the new principle. It seemed that a 30-GeV. machine might be built for the same cost as a 10-GeV. cosmotron type of machine. It is interesting to note that at this time, and quite unknown to the Western world, Russian scientists were planning a scaled-up version of the Berkeley bevatron for 10-GeV.

In December 1952 the Proton Synchrotron Group was formed to augment the original staff of three, and the members of this group, most of them honorary part-time members, remained in their own laboratories and institutes. Members of the theoretical section worked at the Atomic Energy Research Establishment, Harwell, and at the Universitetets Institut for Teoretisk Fysik, Copenhagen. The magnet studies were concentrated in the Laboratoire de Radioélectricité de l'Université, Paris. The radio-frequency problems were studied at the Institut für Physik, Max-Planck-Institut, Heidelberg. The radiation shielding problems at such high energies were worked out using cosmic ray data at the Physikalisches Institut der Universität, Freiburg-i.B., Germany, and the general engineering problems remained at the Chr. Michelsens Institutt, Bergen, Norway, where for six months two of the senior staff of the cosmotron, J. P. Blewett and M. H. Blewett, gave invaluable assistance with the general design problems of the new machine.

By October 1953 enough was known about the implications of the new idea to present a tentative design of an accelerator to the Council of CERN. In America, meantime, the Brookhaven group and another at the Massachusetts Institute of Technology, Boston, had been working on similar problems. At the end of October a conference was held at the Institute of Physics of the University of Geneva, to which were invited representatives of the American groups, the

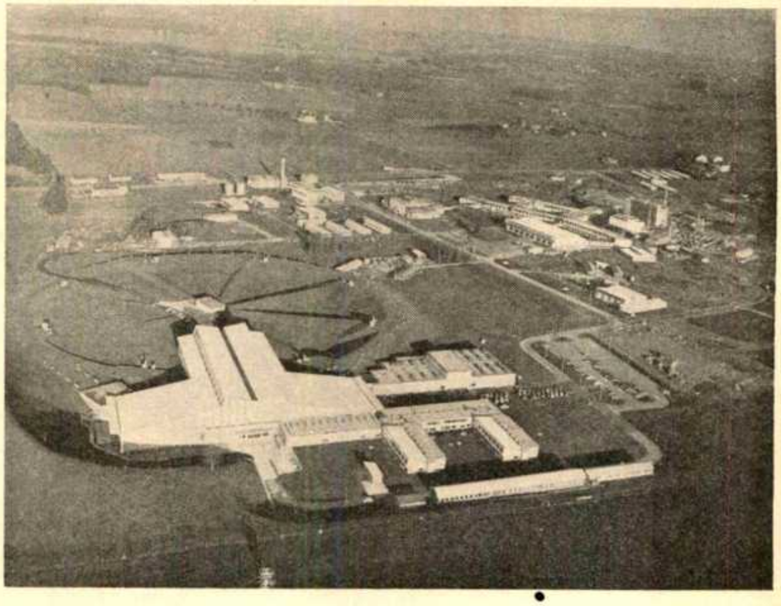

Fig. 1. Aerial view of the CERN proton synchrotron

Table 1

End of year	Main activities	Total staff*
1953	Preliminary studies	20
1954	Design of final machine	75
1955	Development of component parts	120
1956	Major contracts placed for components	150
1957	Manufacturing of components in firms	163
1958	Assembly of components at CERN	176
1959	Testing of components and commissioning of CPS	180

* This includes all staff directly connected with the design and construction of the machine

scientific members of the Council of CERN and other European scientists. The CERN Proton Synchrotron Group gave a series of lectures on its work of the previous ten months and presented a design for a 30-GeV. proton synchrotron using the new alternating gradient principle. It was found that the Brookhaven group had arrived independently at a very similar design.

Immediately after this conference the CERN Council fixed the energy of the machine at 25 GeV. and agreed that the members of the Proton Synchrotron Group should come together in Geneva, near which city the new European laboratory was to be sited. The State of Geneva and the University generously offered temporary laboratory accommodation, and by December 1953 most of the group had moved to Geneva with their families, and the task of designing the final machine started. Soon after this move Dr. O. Dahl resigned as director of the group, and the group suffered a further loss by the sudden death of Dr. F. Goward, who had been acting as deputy director. Despite these initial set-backs the group successfully established itself in Geneva. From the beginning of 1954 onwards staff was steadily recruited from all over Europe, and the group was built up as shown in Table 1.

The alternating gradient principle has been described many times. The original idea was published by E. D. Courant, M. S. Livingston and H. S. Snyder[2] and, quite unknown to these authors, was previously invented by N. Christofilos. In the conventional synchrotron the focusing of the circulating particles is achieved by arranging that the magnetic field which guides the particles around a fixed radius decreases slightly with radius. The amount of focusing achieved in this way is not very great, and consequently the amplitudes of the free oscillation of the particles are large and the dimensions of the magnet gap correspondingly large. Most of the cost in the original synchrotrons is in the magnet; for example, the Russian 10-GeV. synchrophasotron magnet, the radius of which is 30·5 m., contains 35,000 tons of steel and the magnet gap is 150 cm. wide and 40 cm. high. Although greater focusing can be obtained in the axial direction by increasing the gradient of the magnetic field, the criterion for radial stability is then violated. It is the great advantage of the alternating gradient principle that the focusing of the circulating particles can, in theory, be made as strong as one wishes and the amplitudes of oscillation of the particles as small as desired. This is achieved by alternating the sign of the gradient of the magnetic field many times around the circumference of the machine. The net result of this stronger focusing is to reduce greatly the weight of the magnet; for example, the CERN 25-GeV. proton synchrotron magnet has a useful aperture of 14 cm. × 7 cm. and contains only 3,400 tons of steel. It is 100 m. in radius, that is, three times the radius of the Russian machine and one-tenth the steel weight.

These attractive advantages are realized at the price of greatly increased accuracy in manufacturing and aligning the magnet of the synchrotron. Also, once the magnet is set up with the required precision, it must remain in place and, for a machine 200 m. in diameter, this presents some interesting foundation problems. The CERN proton synchrotron magnet is made up of 100 supposedly identical magnet units, each about 4·3 m. long. The magnetic field error between these units for the same energizing current should not exceed about one part per thousand, and the units should be set up with a precision of a few parts per million (0·3 mm. in 100 m. radius). The units are mounted on a free-floating monolith ring of concrete 200 m. in diameter and 2 m. × 2 m. in cross-section, and the machine is built in a subterranean annular tunnel in which the temperature is controlled to ± 1° C. As a further precaution, the concrete ring has steel pipes cast in it, and water is passed round the ring to prevent any part assuming a temperature different from the rest. The technology developed for the magnet construction and the foundations has been described in various CERN reports[3,4].

Another advantage of the alternating gradient principle is that the amplitude of momentum oscillations is very much smaller than in the conventional synchrotron. Since these momentum oscillations result in radial movements of the particles which have to be contained within the width of the magnet aperture, the alternating gradient synchrotron is again more economical in magnet aperture than the conventional synchrotron, in this case by a factor of about 60. Unfortunately, this improvement is also realized at the cost of imposing problems which, at the time of designing the CERN proton synchrotron, seemed formidable.

One restriction concerns the curious behaviour of the machine at an energy called the 'transition energy'. During the momentum oscillations in a synchrotron, particles with greater momentum than the mean value swing out to a radius greater than the mean radius. The revolution time of these particles depends on two factors which work in opposition. Because these particles have a greater momentum than the mean value, their velocity is greater than the mean velocity, and they take a shorter time to complete a revolution than the mean revolution time. On the other hand, since the radius at which these particles circulate is greater than the mean radius, they have a longer path to go round and this lengthens their revolution time. In an ordinary synchrotron, the 'radius term' is greater than the 'velocity term' and those particles with greater momentum than the mean value always take longer to go round than the mean particles. Due to the momentum compaction in an alternating gradient synchrotron, the radial displacement for a given momentum change is about a factor of 60 less than in a conventional synchrotron, with the result that at low energies, at the beginning of the acceleration cycle, the velocity term is larger than the radius term. However, as the energy increases and the particles approach the velocity of light, the velocity term decreases due to relativistic effects. Consequently, there comes a time in the acceleration cycle of the CERN proton synchrotron when the radius term equals the velocity term. The energy at this time is called the 'transition energy'.

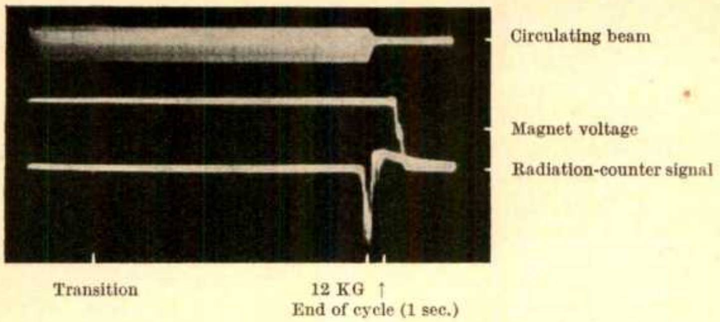

Fig. 2. Beam current versus time. Beam accelerated to about 24 GeV. with no loss

At this energy the frequency of the synchrotron oscillation goes to zero, phase stability is lost, and a rapid shift in the phase of the accelerating voltage has to be made so as to recapture after transition energy all the particles that have been accelerated up to that energy.

It is possible to design an alternating gradient machine so that the transition energy is higher than the maximum energy of the machine, but this can only be done by increasing the circumference of the machine by as much as 50 per cent with a consequent increase in cost. In fact, two new Russian alternating gradient machines have been designed in this way. However, in the case of the CERN proton synchrotron, it was calculated that the transition energy could be passed without loss of particles, and the operational results shown in Fig. 2 justify these calculations. It is possible that the Russian machines will now be modified and consequently will be capable of going to even higher energies.

The particles in a synchrotron circulate at a fixed mean radius, and in order to keep them in the centre of the vacuum chamber during the whole acceleration cycle the frequency of the accelerating voltage must track the steadily increasing magnetic field with high precision. The relative frequency error for a 1-cm. displacement of the mean radius of the circulating particles in the CERN proton synchrotron is given in Table 2.

Table 2

Kinetic energy of protons	$\Delta f/f$ for 1 cm. radial displacement
50 MeV. (injection)	$3 \cdot 5 \times 10^{-3}$
1 GeV.	$8 \cdot 3 \times 10^{-4}$
2 GeV.	$2 \cdot 7 \times 10^{-4}$
3 GeV.	$1 \cdot 2 \times 10^{-4}$
4 GeV.	$4 \cdot 5 \times 10^{-5}$
5 GeV. (transition)	0
10 GeV.	$7 \cdot 0 \times 10^{-5}$
25 GeV.	$7 \cdot 5 \times 10^{-5}$

It can be seen that another unfortunate effect of the transition energy is to impose impossible tolerances on the accelerating voltage frequency at and near that energy. This problem has been solved in the synchrotron in the following manner. From the injection energy of 50 MeV. to about 1 GeV. the accelerating voltage frequency is automatically determined from the magnetic field by an analogue computer having an accuracy of about one part in a thousand. At an energy very much less than 1 GeV., in the 100-MeV. region when the particles are firmly captured in phase-stable bunches, another frequency-controlled system is switched in supplementing the analogue computer. The second system uses pick-up electrodes to detect the phase of the bunches in the machine relative to the phase of the accelerating voltage frequency and, by means of a servo system, holds this phase difference constant at a prescribed value. In addition, the radial position of the circulating bunches of particles in the machine is detected by other pick-up electrodes and, by means of another servo system, the bunches are maintained in the centre of the vacuum chamber.

Apart from these specific problems peculiar to alternating gradient synchrotrons, there are general theoretical problems concerned with non-linear stability criteria which have been studied extensively. The CERN proton synchrotron is equipped with sextupole and octupole lenses to adjust non-linearities in the magnetic field of the machine. A complete description of the CERN proton synchrotron is in course of preparation[3,5,6].

Measurements so far carried out on the CERN proton synchrotron are necessarily preliminary and incomplete. It will take at least six months of measurement work before sufficient is known about the behaviour of the machine to exploit it as a working nuclear physics tool.

The intensity of the proton beam is surprisingly high at this early stage of operation. Protons are injected into the synchrotron at an energy of 50 MeV. from a proton linear accelerator the design of which closely follows that developed at the Lawrence Radiation Laboratory, Berkeley. One important difference, however, is that the CERN proton synchrotron linear accelerator contains magnetic quadrupole alternating gradient focusing in the drift tubes in place of the grid focusing originally used in the American machine. The CERN proton synchrotron linear accelerator has produced intensities up to 5 m.amp. peak although for the tests so far carried out with the synchrotron a collimated 1 m.amp. proton beam has been employed. A buncher and debuncher have been built for this linear accelerator, but neither has been used so far. With 1 m.amp. being injected into the synchrotron and single-turn injection, a capture efficiency of 20–25 per cent has been measured, corresponding to a circulating beam of 10^{10} protons per pulse. The pulse repetition rate at 25 GeV. is 20 per min.

The energy of the protons in the synchrotron has, so far, been calculated from the magnetic field at the time the beam disappears and the radius of the machine. The earliest operational runs were carried out without any of the correcting devices being employed, except for the self-powered pole face windings needed to correct eddy currents in the metal vacuum chamber at injection when the guiding magnetic field is only 140 gauss. The proton beam then disappeared at a magnetic field of about 12 k.gauss due to the number of free oscillations of the particles per revolution becoming an integer. As the magnet yoke becomes saturated, the focusing forces diminish slightly, and instead of the machine working in the stable region between the unstable resonance bands, the operating point is slowly forced into a resonance and the particles are lost to the walls of the vacuum chamber. In later runs the pole face windings were energized by programmed generators designed to keep the focusing forces constant up to magnetic fields of more than 14,000 gauss and it was observed that all the proton beam then reached an energy of just a little more than 28 GeV., limited only by the peak magnetic field available in the CERN proton synchrotron.

An attempt has been made to measure the number of free oscillations per revolution in both the axial and radial directions (Q_V and Q_R) during the acceleration cycle. At injection, with the pulsed inflector voltages not energized and a 2-μsec. pulse of protons injected from the linear accelerator, the beam spirals inwards in radius due to the rising magnetic field for over one hundred microseconds, and the fractional Q-values can be observed in the two directions by pick-up electrodes sensitive to one or the other of the two directions of oscillation. For these measurements the beam is injected in such a way as to set up either axial or radial oscillations of the particles. During the acceleration cycle the Q-values were measured by pulsing the quadrupole correcting lenses at different times in the cycle and by variable amounts, and noting when the beam was lost. The nominal Q-values are both about 6·25, and if at a certain moment the quadrupole lenses are pulsed by an amount that shifts, say, the Q_R value to 6·0, the beam will be lost due to instability at a radial first resonance. Similarly, pulsing the lenses in the opposite direction by the same amount loses the beam due to instability at a radial second-order resonance at $Q_R = 6{\cdot}5$. Apart from the shift of the Q-values when the magnet becomes saturated, it appears that they remain close to $Q_V = 6{\cdot}3$, $Q_R = 6{\cdot}2$ during most of the acceleration cycle. Using this same method, the unstable boundaries of the stable operating region, namely $Q_R = 6{\cdot}0$, $6{\cdot}5$ and $Q_V = 6{\cdot}0$, $6{\cdot}5$, have been measured. So far, no higher-order resonances due to non-linear instabilities have been observed, although whether this is a tribute to the linearity of the machine or due to the crudeness of the measurements remains to be seen.

Measurements have been made of the closed orbit displacement at 20 points around the circumference of the machine, from which the closed orbit amplitude and shape have been computed. The closed orbit is that orbit around which all particles perform free oscillations. In a perfect machine without field-free sectors it would be a perfect circle in a plane. In an imperfect machine its shape and amplitude are due to the imperfections. At injection the peak-to-peak amplitude of the closed orbit in the radial direction is about 4 cm., and in the axial direction it is a few mm. Later on in the cycle the peak-to-peak amplitude diminishes in the radial direction to about 1 cm. and remains negligible in the axial direction. Since the vacuum chamber dimensions are 14 cm. in the radial direction and 7 cm. in the axial direction, there is therefore no danger of loss of beam due to magnet misalignments.

The transition energy, which seemed so formidable a barrier during the design stage, proved to be easily surmountable in practice. The precision of phase switching needed at transition for no noticeable loss of particles was found to be about ± 3 msec., or ± 36 gauss.

Both the computer and the beam control system for determining the frequency of the accelerating voltage have proved very satisfactory after minor adjustments. During the operational runs so far carried out the beam control system has been switched in about 1 msec. after injection and there seems to be no noticeable loss of particles during this switch-over. During the early acceleration studies the radial control servo acted on the amplitude of the accelerating voltage, which in principle is equally effective a means of controlling the radial position of the beam as varying the phase or frequency of the accelerating voltage. This arrangement was not successful, perhaps due to a certain amount of jitter in the phase-lock loop, and the system was modified so that the radial control servo acted on the phase of the accelerating voltage. This second method was completely successful and no particles were lost during acceleration. Preliminary measurements indicated that the beam was not moving more than 1 cm. from the centre of the vacuum chamber during the acceleration cycle. The beam control system depends for its operation on the presence of a bunched proton beam, and due to noise in the servo loops there is a certain minimum value for the circulating beam current below which the system no longer maintains control. It appears from early measurements that the system will work with about 10^8 protons per pulse.

Table 3

Accelerator	Max. energy (GeV)	Mean intensity (particles per sec.)	Completion date
Brookhaven proton synchrotron (Cosmotron)	3	2×10^{10}	1952
Saclay proton synchrotron (Saturne)	3	10^{10}	1958
Princeton–Pennsylvania proton synchrotron	3	2×10^{11}*	1960
Berkeley proton synchrotron (Bevatron)	6	2×10^{10}	1954
Rutherford Laboratory proton synchrotron (Nimrod)	7	10^{12}*	1961/62
Russian A.G. proton synchrotron	7	$\sim 2 \times 10^{9}$*	1960
Russian proton synchrotron (Synchrophasotron)	10	$\sim 10^{9}$	1957
Australian proton synchrotron	10	10^{7}*	1962/63
Argonne zero gradient proton synchrotron	12·5	2×10^{12}*	1962
CERN proton synchrotron	28 25 6–10	2×10^{9} 3×10^{9} 10^{10}	1959
Brookhaven A.G. proton synchrotron	30	$\sim 3 \times 10^{9}$*	1960
Russian A.G. proton synchrotron	50	$\sim 10^{9}$*	1961/62

* Target figures.

A very important parameter of an accelerator is the mean intensity. Table 3 shows that at an energy of 6 GeV. the mean intensity of the CERN proton synchrotron is now nearly as high as that of the bevatron, and at 10 GeV. considerably higher than that of the Russian machine. It is hoped to increase the intensity of the CERN proton synchrotron by an order of magnitude by bringing into operation the buncher and debuncher of the linear accelerator and allowing more protons to be injected into the synchrotron. At 25 GeV. the yield of secondary particles from internal targets is considerably higher than at 6–10 GeV., and early measurements of secondary particle yields from the synchrotron indicate that the machine may be a strong competitor to the lower-energy high-intensity machines now being built from this point of view.

The significance of this accelerating machine can best be understood in relation to other proton synchrotrons. The CERN proton synchrotron accelerates protons to an energy which is about three times the energy of the largest accelerator now functioning, namely the Russian synchrophasotron

Table 4

	Maximum energy	Cost in million (Swiss francs)
CERN proton synchrotron	28 GeV.	120
Rutherford Laboratory proton synchrotron	7 GeV.	84
Saclay proton synchrotron	3 GeV.	53
Hamburg electron synchrotron	6 GeV.	60

at Dubna, and so, for a while, Europe possesses by far the largest accelerator in the world. In 1960 another machine of slightly higher energy (30 GeV.) will come into operation at Brookhaven in the United States, and in 1962 or thereabouts the Russians plan to bring into operation a machine twice as large as the CERN proton synchrotron (50 GeV.). The possession of the world's highest-energy accelerator is, and always has been, a transitory state, but the remarkable significance of this event is that for the first time for more than twenty years Europe can claim this possession. Since the War a major effort has been made in Europe to equip laboratories working in high-energy nuclear physics research with the necessary apparatus. The magnitude of this effort in terms of money is considerable, as can be seen in Table 4.

The cost of running these laboratories and building apparatus for the experiments is equally high, and this is a recurrent cost since new experimental apparatus is continuously being needed as the experiments develop. For example, the annual budget of CERN, which in addition to the CERN proton synchrotron also has a 600-MeV. synchro-cyclotron, is about 60 million Swiss francs per annum.

That Europe has made such an effort in this field of fundamental research has been due to the foresight of the physicists who have planned the laboratories and the sympathy of the national treasuries which have supplied the money.

[1] Adams, J. B., *Nature*, 184, 944 (1959).

[2] Courant, E. D., Livingston, M. S., and Snyder, H. S., *Phys. Rev*, 88, 1190 (1952).

[3] Regenstreif, E., CERN 59-29. "The CERN Proton Synchrotron". Part 1. Ch. 1: Basic Principles of Alternating Gradient Synchrotrons. Ch. 2: Design of the CPS and Choice of Parameters. Ch. 3: Magnet. Ch. 4: The Radio Frequency Accelerating System.

[4] Adams, J. B., CERN 56-21. "The Design of the Foundations for the Magnet of the CERN Alternating Gradient Proton Synchrotron",

[5] Regenstreif, E., CERN 59-26. "Le Synchrotron à Protons du CERN, 2 ème partie. Ch. 5: L'injection des particules" (English version to be published).

[6] Regenstreif, E., CERN. "The CERN Proton Synchrotron", Part 3 (in the press).

HUMAN ADAPTATION TO COLD

By Dr. LAURENCE IRVING

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THE people from whom we have gained our opinions about physiological reactions to cold have lived in clothing manufactured to common standards and in shelter of regulated warmth. Until recently physiologists have not been impressed by the significance of man's adaptability to cold. But we seem to forget that in every military campaign some soldiers have learned to perform well during prolonged exposure to cold which would have been unbearable in light clothing and poor shelter unless they had developed the adaptability that remains latent without training.

Recognizing this, Scholander *et al.* started expeditionary physiological studies of populations motivated and trained for exposure, including young Norwegians briefly trained in the cold[1] and three different ethnic groups; Arctic Lapps[2], Australian Aborigines[3] and arctic Athabaskan Indians[4]. The results have demonstrated an unexpected range and variety of reaction to cold among people living in cold or chilly climates.

If development of adaptation to cold is associated in our minds with inability to obtain what we regard to be proper protection, we would disregard the many religious individuals and others that have elected to go in thin clothing with bare feet to practise humility and escape from the distraction caused by indulgence for bodily comfort.

The Alaskan members of the Wisdom, Knowledge, Faith, Love Fountain of the World commonly wear only a cotton robe and go with heads, hands and feet bare. They keep their clothing light out of regard for simplicity; but it is their opinion that their garb is more comfortable than the designs of fashion. They avoid endangering themselves by their light clothing, for when the ground is too stony they put on shoes and they add further protection in severe cold weather.

Two young men, students at the University of Alaska, who have been members of the Fount at Venta, Alaska, for about two years, co-operated in measurements of their skin temperature. One of the students, *B*, stood for 64 min. in a room at an air temperature about 0° C., barefoot on a wood floor at − 1° C. to + 1° C., clad in shorts with a loose short-sleeved cotton robe. Towards the end he began to shiver slightly and at 42 min. the side of one big toe had cooled to 7·9° C., thereafter warming to 18·5° C. at 53 min. and cooling again to 15° C. at 64 min. At 17 min., when he remarked feeling both big toes warming, the recorded temperature on one toe had risen from a momentary low at 10·0° to 10·2° C. in the preceding minute and rose in the next minute to 10·4° C. At 34 min. the big toe had cooled again to 8·8° C., a change that he also reported as it began. At 42 min., when the toe was 7·9° C., it felt uncomfortable but not painful. 30 sec. later, when the toe had warmed to 8·6° C., he had already informed me of the warming. His body felt uncomfortably chilly, and he directed attention to sweating in the armpits which he said often accompanied reaction to chill.

The second student, *G*, stood in the cold room (air 1° C. to 2° C., floor − 2° C. to − 3° C.) for 34 min. His big toe cooled to 4·9° C. in 20 min. and to 4·6° C. in 34 min. At 20 min. his legs shivered in vigorous,

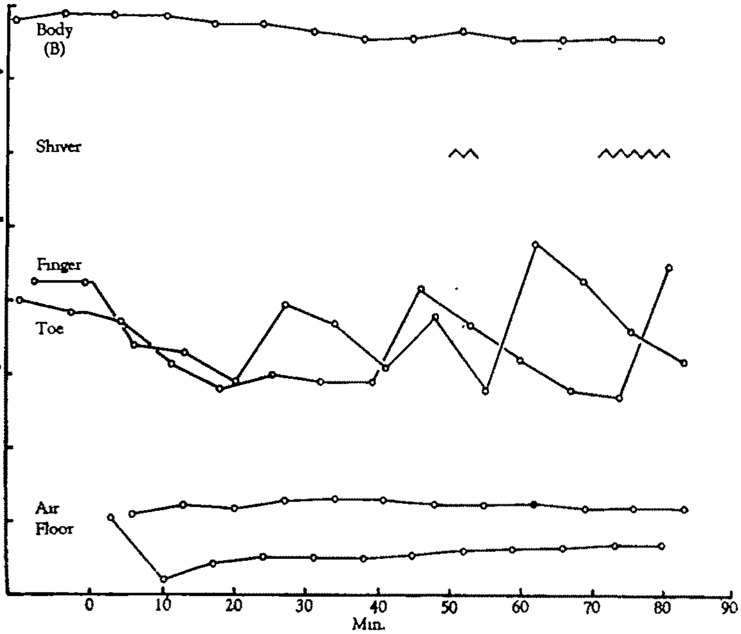

Fig. 1. Record of skin temperature in student *B*. Note the large swing of temperature in fingers and toes and the very slight shivering

intermittent bouts and he reported that his body felt warm and that his feet were not painfully cold. The test was not terminated by cold but owing to dizziness from standing.

On a second occasion, temperatures were recorded with the students sitting reading in the cold room, dressed as before. The record in Fig. 1 is for student *B*, and shows the average reading from two thermocouples on the back and one on the sternum (the three not differing more than 1·5° C.), one on the mid-finger pad and one on the big toe near the base of the nail. The tip of the middle toe held within 2° C. of the big toe. The outer side of the foot 2 cm. from the base of the little toe and the dorsum were about 10° C. warmer than the toes. Legs were intermediate between the temperatures of the body and feet.

The finger of student *B* began to warm and cool in long cycles after 20 min. (Fig. 1), his toes warmed at 40 min., and their warming at 60 min. was asynchronous with the warming of the finger. Student *G*, who shivered somewhat more but not vigorously, showed no clear-cut warming of the toes but a distinct warming of the finger. Although his toes were colder than 5° C. from time 65 to 100 min. they were not painful. The reactions to painful cold are usually so evident that it is unlikely that they could be deliberately suppressed and the students located and described their other sensations very accurately. Both students said that although they would not choose such conditions for study the cold did not interfere with their reading. Airman Henson, who assisted in the observations, and I were chilly in warm clothing as we moved in and out of the cold room.

A contrasting response was shown by the airman, who had been in Alaska for about two years. Sitting with bare head, hands and feet, and wearing only cotton fatigue jacket and trousers over shorts, about equivalent in insulation to the robes of the two students, his fingers and toes did not cool quite so low as those of the students (Fig. 2). He began shivering almost at once and continued in violent intermittent bouts which soon reached disagreeable intensity. After 8 min. his toes felt very uncomfortable and were very painful after 14 min. At 41 min. it was considered that his fortitude was not contributing further to physiology, and he reluctantly consented to terminate the test.

In spite of his shivering, which perhaps cooled him by ventilating his clothing as much as it contributed in metabolic heat, the airman's body skin cooled promptly. The two students had kept their bodies comfortably warm with but little shivering. As student *G*'s toes cooled to about 5° C. at 65 min., he remarked that they lost sensation and he could not detect a light touch with a pencil. The feet of all subjects were flushed and somewhat swollen, blanched on pressure and on release immediately reddened again. Spontaneous blanching of the feet was not observed, and judging by this criterion frost-bite was not imminent. After leaving the cold room the students re-warmed promptly with no pain; the airman took about 15 min. to regain comfortable warmth, with diminishing pain as recovery proceeded.

Although one of the students lost sensitivity in his toes when they cooled to about 5° C., otherwise both of them acutely observed their sensations of cold and warmth and located them with surprising accuracy, which surprised me and the airman. The students reported and located warming of even one toe by a fraction of a degree and accurately named the cessation of the warming phase of the cycles.

Student *B* pointed out that visible beads of sweat formed on his finger tips as they began to warm and showed me sweat beads on his warming toes after the tape covering the thermocouples was removed.

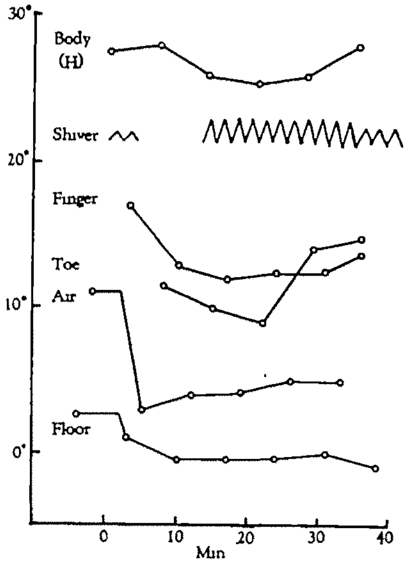

Fig. 2. Records of skin temperature in Airman Henson. Violent shivering with considerable fall of surface temperature

The airman also pointed out sweat on his still painfully cold toe as it warmed after the thermocouple was removed. Localized sweating thus appears to be connected with the vascular reaction which brings about warming of cold skin. It appears that local re-warming is associated with sweating. Sweating seems inconsistent with very cold skin; but the surprising combination may furnish a clue to the nervous component involved in peripheral regulation of temperature.

These adapted individuals appeared to have developed accurate appreciation for the temperature of their skin, keeping track of the changes at various locations at all temperatures down to about 6° C. They apparently deliberately utilize this conscious scanning of the thermal situation to avoid unsafe exposure. Eskimos also observe and care for the thermal situation in their faces and hands in extreme cold, and rarely experience frost-bite except through accident or when required by circumstances deliberately to tolerate the well-known sensations which warn of incipient frost-bite.

The freedom of the adapted men from pain caused by cold was associated with retention of adequate sensitivity and probably with improvement in the recognition of temperature of skin, its small changes and exact location. Keen appreciation of thermal conditions must be employed for the maintenance of skin to be safe at temperatures critically close to freezing, for, like hibernating animals, very cold tissues lack heat and the power for its production which buffer warm tissues against the fatal consequences of small depressions in temperature. I have remarked that sensitivity persists in the very cold feet of arctic birds and mammals which are still accurately guided by the sensations requisite for standing and movement[5]. Only sensitive feet can safely serve to economize on heat by cooling to nearly freezing, and it is obvious that insensitive tissues could perform no adaptive function.

It is also essential that any surface of an animal normally making contact with cold snow or ice should not become warm enough to cause melting or the skin or hairs would quickly freeze fast[6]. The students said that when they stepped from a warm room on to cold icy ground their feet became iced and picked up gravel, causing painful abrasions, although if icing did not occur their feet could withstand even − 20° C. for a time. Those who have hunted with dogs in cold snow will recall that the feet of some kinds become crippled by icing. The feet of healthy arctic animals do not commonly become iced as long as they are free to care for themselves.

After habitually wearing shoes, learning to walk with bare feet on rough ground involves some superficial hardening, but the process of learning to utilize the latent adaptability also involves improvement in operation of the senses which must direct the bare feet so that they are guided to avoid violent contacts with sharp or hard objects which no living tissue could withstand. In cold-adapted feet it is evident that the temperature causing cold pain is lowered, but sensitivity to temperature and the other senses necessary for the use of the feet must remain adequate for their effective operation.

These two students have deliberately practised exposure to cold in Alaska for only two years, but their exposure was less frequent and severe than among some of their people who worked out of doors. The airman appraised the practical importance of their adaptation by remarking on the advantageous mobility that would be gained by an infantry battalion trained to operate in such light clothing; but when I remarked that airmen would gain equally if relieved from some of the impediments of clothing and shelter he was less pleased with the idea.

It is apparent that human adaptability to cold is of practical importance when it is evoked by careful training motivated by consideration of benefit to be obtained from exposure. It is also apparent that adaptability to cold remains a latent faculty in urban populations.

Dr. Raymond J. Hock made instruments available from the Arctic Aeromedical Laboratory, and generously helped with preparations and part of the observations. His assistant, Airman First Class Harns C. Henson, of the Arctic Aeromedical Laboratory, gave skilled help throughout the experiments.

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[1] Scholander, P. F., Hammel, H. T., Andersen, K. L., and Løyning, Y., *J. App. Physiol.*, 12, 1 (1958).

[2] Andersen, K. L., Bolstad, A., Folkow, J. B., Fox, R. H., Jorgensen, A., Krog, J., Løyning, Y., Nelms, J. D., and Wilson, O., *J. App. Physiol.* (in the press).

[3] Scholander, P. F., Hammel, H. T., Hart, J. S., LeMessurier, D. H., and Steen, J., *J. App. Physiol.*, 13, 213 (1958).

[4] Irving, L., Andersen, K. L., Bolstad, A., Elsner, R. W., Hildes, J. A., Løyning, Y., Nelms, J. D., Peyton, L. J., and Whaley, R. D. (in preparation).

[5] Irving, L., and Krog, J., *J. App. Physiol.*, 7 (4), 355 (1955).

[6] Irving, L., *The Physiologist*, 2, 18 (1959).

COMMITTEE ON NATURAL RESOURCES IN SCOTLAND

By Dr. F. N. WOODWARD, C.B.E.

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A COMMITTEE on Natural Resources in Scotland has been set up by the Scottish Council (Development and Industry) under the chairmanship of Mr. L. A. Elgood to examine the most suitable ways of developing and conserving Scottish primary resources. At the outset, two stages are envisaged: first, the setting-up of a fact-finding study group to provide information and, secondly, a subsequent conference of wider educational scope with emphasis on socio-economic problems.

The study group, which will meet at the Royal Society of Edinburgh during October 31–November 2, will comprise about fifty invited national and international experts in the various resource fields, who will discuss and review previously prepared papers designed to cover these problems in three phases, namely, individual resources already surveyed, the inter-relationship between them, and resource administration techniques and development.

The published proceedings of this fact-finding study group will not only make available to the public for the first time an up-to-date assessment of Scotland's primary resources, but also provide the background information for the subsequent more comprehensive conference which is intended for a larger, less technical audience, and would be designed to influence public opinion.

It is anticipated that the complete inquiry will cover a period of 3–4 years and inevitably lead to a change in the public's attitude towards problems of resources.

While it is not possible to anticipate the outcome, it is envisaged that the need for additional research institutions and for a permanent centre for information and for the co-ordination of the interests and efforts of those concerned with conservation and use of resources will emerge.

The setting up of the Committee on Natural Resources in Scotland is a logical if belated outcome of the United Nations Scientific Conference on the Conservation and Utilization of Resources which was held at the United Nations temporary headquarters at Lake Success in 1949. That Conference was unique among international conferences in that it provided a forum for more than six hundred scientists, conservationists and economists concerned with assessment, conservation and development of resources, drawn from fifty different countries. Their deliberations, in which the large British and Commonwealth delegation under the leadership of Sir Harold Hartley took an active part, in the presence of 350 observers resulted, among other things, in the production of seven volumes of "Proceedings" which are now recognized as the classic reference authority on world resources.

Among many suggestions emanating from the Conference was one to the effect that all countries should be regarded as under-developed, and that their known and potential primary resources should be examined afresh. This has been followed by resource surveys and conferences in various parts of the world. In the United Kingdom, however, there has been no comprehensive conference to follow the example set by the United Nations Conference. There are many reasons why such a conference would be difficult to organize, and even if it were, the outcome would be of debatable value ; but Scotland, on the other hand, presents a sufficiently compact region and has, at the same time, the diversity of resources to yield good results from such an inquiry. These resources are soil, water, minerals, fuel, fish and wild-life, forestry, and people of particularly interesting character.

A persistent problem in regard to the last of these is the continuous drainage of Scottish people through emigration. At present, about 40 per cent of Scottish graduates in science, and 60 per cent in engineering, leave to work elsewhere. While there are many examples of a flow in the reverse direction, the overall loss by emigration of trained men is too high and is a matter for serious concern.

There are other obvious resource problems awaiting solution in Scotland, and many more will doubtless be revealed as the inquiry develops.

For example, peat covers 1·7 million acres of land : if technical problems now under investigation by the Scottish Peat Committee are solved, a new industry could be established using an indigenous natural resource, with coincidental land reclamation for productive use. The conflict between agriculture, forestry and local authorities for land-use is being energetically tackled by the Scottish Departments and others : the attack on ancillary problems—such as the infestation by bracken of more than 0·5 million acres of land and the industrial use of agricultural crops and by-products—on the other hand, has been sporadic and frequently uninspired.

Although the demand for water is increasing rapidly, no single agency exists to survey and regulate water resources. There is a lack of hydrometric and underground water data and no standard technique for river flow gauging.

Also, although Scotland is very well surveyed geologically, very few geophysical surveys have been undertaken and no facilities exist for mineral beneficiation, research and development.

Due to historical trends, certain of the British natural resources agencies operate entirely within the Scottish regional context, whereas others have an overall United Kingdom interest. For example, fuel supply and forestry are dealt with on a United Kingdom basis, whereas agriculture is administered from a central Scottish headquarters ; water is not the concern of any particular agency.

Scotland's natural resource endowments, although significant, are relatively limited in number. Some of these, such as coal, oil-bearing shale, and water for hydro-electric power, have already been exploited, and others can, through research, form a source of wealth in the future.

In this respect, Scotland is handicapped in that its Government-supported resource laboratories, besides being small in number, are in imbalance. Though there are eleven concerned basically with the use of soil for agriculture, not one is solely devoted to forestry. The only other primary resource laboratories in Scotland are those dealing with fish, jute, peat, coal and nature conservancy ; none, for example, is available for undertaking research on mineral beneficiation, non-fish marine resources or chemurgy.

A good example of primary resource development is to be found in the new seaweed-based industry in Scotland which resulted from the co-operation between industry and an independent research organization specifically set up for the purpose.

Quite apart from the lead given by the United Nations Conference, it has been felt for some time that a new and integrated study of Scottish resources —including man—should be undertaken on the widest possible scale.

It is realized, of course, that such inquiry could result in recommendations which might affect certain present-day economic interests, although the ultimate result would be in the national interest.

The immediate use of peat for fuel, for example, might conceivably react to the detriment of future generations whose need for peat for water-storage purposes might be more vital. On the other hand, the locking of certain resources would not necessarily be a good thing, since unforeseeable changes, resulting from advances in scientific knowledge, could in the future make such action futile.

It is the hope of its promoters, however, that, among other things, the inquiry may be instrumental in preventing development of one resource from permanently damaging another, and thus avoid the chaos which has resulted in many parts of the world from haphazard and unco-ordinated development.

The Secretary of the Committee on Natural Resources in Scotland is Mr. H. M. Wilson, The Scottish Council (Development and Industry), Edinburgh.

OBITUARIES

Prof. Eric S. Horning

On November 14, 1959, British cancer research lost one of its prominent figures by the death of Dr. E. S. Horning, professor of experimental pathology in the University of London at the Chester Beatty Research Institute. Australian by birth, he served as a volunteer with the Imperial Forces during the First World War, and went to Oxford in 1920 to start his professional career in the honours school of comparative anatomy and zoology.

Having taken his degree, he went into research, first in Melbourne with Prof. Agar and later with A. Fischer in Berlin-Dahlem and E. V. Cowdry and A. Carrel in the United States. From this period stem numerous papers dealing with the behaviour of mitochondria and other cytological problems. The excellence of his early work was soon recognized. In 1932 he contributed by invitation an article on the enzymatic function of mitochondria to "Ergebnisse der Enzymforschung", and in 1941 he wrote in G. Bourne: "Cytology and Cell Physiology" the chapter on micro-incineration and the inorganic constituents of cells, a subject on which he was an authority.

Armed with a comprehensive knowledge of cytology and histology, and well versed in the methods of tissue culture, in 1933 he returned to England, the country he loved more than any other, where he spent the rest of his life but for the years of the Second World War, during which he served again with great distinction in the Armed Forces.

In 1933 Dr. Horning joined the staff of the Imperial Cancer Research Fund as a Beit Fellow; when six years later the outbreak of war called him from the bench he had made his name as a cancer investigator. A series of brilliant papers had appeared in which Cramer and Horning recorded their original observations, shedding new light on the pathogenesis of the tumours of the breast, pituitary and thymus, and establishing the role of the adrenals in the development of these tumours.

In 1947 Dr. Horning accepted an invitation by Prof. A. Haddow to go to the Chester Beatty Research Institute as reader in experimental pathology. There he worked until the end, his scientific activities continuing unabated in spite of periods of indifferent health. Testimony to his persistent interest in the endocrinological aspects of cancer are the chapters in "Oestrogens and Neoplasia", his contribution to "Causation of Cancer" in the *British Medical Bulletin* 1958, and his papers on the experimental cancers of the prostate and the œstrogen-induced tumours of the kidney. Although from 1936 onward the role of hormones in neoplasia was the main theme of his endeavours, by his studies of the action of carcinogenic compounds on grafted tissues he made notable contributions to chemical carcinogenesis. Besides, his considerable experience with the pathology of the experimental and spontaneous tumours of rodents made him an invaluable collaborator in many projects of the Chester Beatty Research Institute. *Inter alia,* he described with Boyland the tumours induced by nitrogen mustards in mice, and with Alexander the reaction of the subcutaneous tissue of the rat to implanted plastic films.

Anybody who had the privilege to visit Dr. Horning in his laboratory and to look at some of his slides could only admire the beauty of his histological preparations, and was no longer astonished by the excellence of the photomicrographs which adorn his papers. The high quality of the material on which Dr. Horning's results were based and the elegance of his techniques rather than ingenuity of speculation were the basis of the high esteem he enjoyed among his colleagues at home and abroad. But, above all, few men had his gift for friendship and a heart so warm and generous. Only those who have worked with him know how easy he made life for his collaborators and how much he did unselfishly to further the work of the Institute. For his human and scientific qualities the memory of Eric Horning will be cherished in many lands.

F. Bielschowsky

Mr. W. E. Dick

With the sudden death of William E. Dick at the early age of forty-five, scientific journalism has lost one of the greatest exponents of this now rapidly expanding and increasingly important field—a field which shows signs of becoming to-day almost as important to the scientist, as a means of keeping him informed of developments in other fields as well as his own, as to the intelligent layman who wishes to learn about scientific and technological progress. He was successively editor of *Discovery* (1945–56), *British Chemical Engineering* (1956–58), and *Chemistry and Industry,* the journal of the Society of Chemistry and Industry. Prior to that he had held posts on the *Grantham Journal* (1941) and the *South Western Star*, which he joined in 1942. For a brief period he then worked as assistant editor to *Chemical Age*, before joining *Discovery* in a full-time capacity in September 1944.

William Dick was born at Walthamstow on July 12, 1914, and was educated at King's School, Wimbledon, where he took a State scholarship at the age of fifteen to the Imperial College of Science and Technology to read biology. Although he left the Imperial College before his degree was completed, his training was not purely journalistic, for he later became a qualified scientist, taking an external B.Sc. degree at the Chelsea Polytechnic in 1942. He was also elected a Fellow of the Linnean Society.

During the War he served with the Auxiliary Fire Service during the first months, and then at the British Expeditionary Force, Royal Army Service Corps Headquarters in France before being evacuated when France surrendered. Then he transferred to one of the camouflage units of the Royal Engineers at Farnham. He entered journalism in August 1941, after being invalided out from the Army.

When Bill Dick became editor of the newly revived *Discovery*, science writing was still, as I was made only too well aware on numerous occasions, regarded as a 'bastard art', more often than not despised by the scientist and badly handled by the Press. By setting a standard of journalistic and scientific reporting that raised *Discovery* from being a relatively unknown journal to one that had a readership throughout the world, and was renowned and highly regarded as a scientific paper, Dick showed and taught others that science writing could be a valued and respected branch of science. In the process he also not merely set a new standard for other people to follow, but also helped to give invaluable advice and training to many new-comers venturing into this field. Besides his activities as editor for *Discovery*, Dick also wrote many articles, one of the best known of which was "Science and the Press", which was written in 1954 for the journal *Impact*. Here he set out many of the problems he had encountered and the ideals in which he believed. He also wrote, or contributed to, a number of books as, for example, "Science and the Welfare of Mankind", written in conjunction with I. B. N. Evans, published by Simpkin Marshall in 1946, and "The Story of Energy", published in 1951 for the Bureau of Current Affairs. In 1957, Butterworths Scientific Publications brought out his "Atomic Energy in Agriculture".

Dick's interests, indeed, were catholic in the extreme, every field of science intrigued and on occasion excited him, although professionally by qualification he was a biologist. The latter fact explains perhaps to some degree his immense interest in flowering plants and the joy and enthusiasm which he brought to his gardening.

Despite his many achievements, William Dick suffered for twenty years from a long series of illnesses of increasing severity. The debt owed him by scientific journalism is immense, and his early passing is an occasion of great sadness. Personally, also, he will be missed as one of the kindest and most generous of people, especially in times of trouble.

DEREK WRAGGE MORLEY

IT is impossible to conceive of any scientist, certainly of any editor or journalist, who was not shocked by the news of Bill Dick's sudden death or who will not now miss his entertaining chatter and stimulating pronouncements on science, especially on the exposition of science.

Although he was initially a reporting journalist and eventually an editor, he was never satisfied merely to sit down and rewrite into more readable form scientific reports and research publications which came to hand. He really believed that the good editor wears out the soles of his shoes rather than the seat of his pants. He went after news personally and wrote it up personally. For this reason alone, it is true that as Mr. Wragge Morley writes: "*Discovery* [metamorphosed] from being a relatively unknown journal to one that had a readership throughout the world, and was renowned and highly regarded as a scientific paper". Though *Discovery* still maintains this excellent standard, I am sure the present editor would be the first to admit that this happy editorial evolution was originally inspired by Bill Dick.

Dick's remarkable achievements and his engaging, though somewhat naïve, personality did not pass unnoticed, especially among his journalistic and editorial colleagues. For example, 1956 saw the tenth anniversary of the new *Discovery* which coincided with the tenth anniversary of Bill Dick's editorship. To mark this double event, a sherry party was held to which only editors and science writers were invited. It was significant that everybody who had been invited attended to pay tribute to Dick and *Discovery*.

Bill had a warm heart and a ready tongue. He had a real gift for nosing out news. His sincerity could never be challenged, for if he admired a person or his achievements he was lavish with praise; but woe betide anybody who, in his opinion, was not playing the game. You always knew how you stood with Bill.

It is a tragedy that science and especially the exposition of science (mainly through popular publications) has now to suffer the loss of this remarkable man; for science still suffers from a dearth of good writers, in spite of the fact that, as Mr. Wragge Morley says: "Science writing [is] a valued and respected branch of science".

The presentation of scientific advancements and achievements in an understandable form has now become an essential branch of science itself. Research cannot go on indefinitely behind closed doors; there must eventually be a fearless exposition and interpretation of the truths it unfolds. Bill Dick believed this and worked selflessly and passionately to this end.

L. J. F. BRIMBLE

Dr. H. P. Wilkins

HUGH PERCIVAL WILKINS, the distinguished selenographer, died at his home at Bexleyheath, in Kent, on January 23.

Wilkins was never a professional astronomer. He was born and educated in Carmarthen, and after serving in the Army during the First World War he became an engineer. His main interest lay, however, in astronomy. Though he made useful planetary observations from time to time, he concentrated almost entirely upon the Moon, and by 1924 had completed a lunar map 60 in. in diameter. Eight years later he commenced work upon an even larger chart, to a scale of 300 in. to the Moon's diameter. A prodigious amount of work went into this project; except during occasional periods of ill-health, Wilkins was nearly always busy at his telescope whenever the sky was clear and the Moon visible. The 300-in. map was published in 1951, and he then undertook a further revision, which appeared in 1954. Yet another revision was planned, though unfortunately he did not live to complete it.

After his marriage, Wilkins moved from Wales to Kent, and set up a 12½-in. reflector, later replaced by a 15¼-in. instrument. It was with this equipment that he carried out most of his work, though he also made numerous observations with very large instruments in both Europe and the United States. He was a regular contributor to many scientific periodicals, and held the position of director of the Lunar Section of the British Astronomical Association from 1946 until his resignation ten years later. In 1941 he gave up practical engineering, and joined the Ministry of Supply, retiring only at the end of 1959 with the intention of spending the last part of his life in astronomical work.

Wilkins was well known as a lecturer on astronomy; he made frequent broadcasts on both sound radio and television, and was the author of seven books, the last of which was completed only a few days before his death. In 1955 the University of Barcelona conferred on him an honorary Ph.D. degree in recognition of his work for selenography.

Wilkins was above all a kindly man; he was unfailingly courteous, and could never understand those who were not. His enthusiasm for astronomy was inspiring, and he was always ready to help others. A deep sense of personal loss will be felt by his many friends all over the world.

PATRICK MOORE

NEWS and VIEWS

Chemical Engineering at Swansea : Prof. E. S. Sellers

Prof. E. S. Sellers resigned his post as head of the Department of Chemical Engineering, University College of Swansea, on January 31, to join the British Petroleum Co. as manager of the Petroleum Division at the Sunbury Research Centre. When in 1955 Prof. Sellers went to Swansea from the Department of Chemical Engineering, University of Cambridge, he faced the task of building a new department in a College that planned unusually rapid extension to its facilities for education in applied science. He leaves a department of chemical engineering with a well-established teaching programme that makes provision for the admission of some thirty honours students annually and the expectation of expansion to an entry of fifty students when new accommodation becomes available during the next five years. A special feature of the course for chemical engineers in Swansea has been the more than usual involvement of local industry. A considerable part of the practical work for undergraduates has been carried out on industrial plant rather than on laboratory equipment in the Department. This has been made possible by the excellent relations that the Department of Chemical Engineering has established with industry. During his period in Swansea, Prof. Sellers has supervised research on the flow of non-Newtonian liquids and the control of continuously operating process plants. In 1957 he was awarded the Moulton Medal of the Institute of Chemical Engineers for work (with Dr. D. R. Augood) on the distillation of hydrogen. He has served the Institution of Chemical Engineers as a member of the Council and on the Board of Examiners. He has been a member of several advisory committees of the Admiralty and the Ministry of Supply.

Prof. J. F. Richardson

Prof. J. F. Richardson, who succeeds Prof. E. S. Sellers at Swansea, graduated at the Imperial College of Science and Technology in 1941. After six years of postgraduate research with Sir Alfred Egerton, he was appointed lecturer and later senior lecturer at the College. In 1958 he resigned his university appointment to join the Development Division of Messrs. A. Boake Roberts and Co., Ltd., as senior chemical engineer. Prof. Richardson has served the Institution of Chemical Engineers for four years as a member of the Board of Examiners, as honorary librarian since 1954, and as a member of the Nominations Committee, the Careers Advisory Committee and the Advisory Panel on student membership. His publications cover a wide range and include a text-book on chemical engineering written jointly with Prof. J. M. Coulson. During 1941–47 his work was largely concerned with laboratory and large-scale studies on fire prevention and extinction. More recent research by Prof. Richardson has related to the physical properties of two-phase systems.

Royal Naval Scientific Service : Mr. J. Buckingham, C.B.

Mr. John Buckingham, who recently retired from the Royal Naval Scientific Service, where he held the post of director of research programmes and planning, joined that Department as a technical assistant in the Board of Invention and Research in 1917 and made naval science his career. During the years between the Wars he played an important part in the development of what was ultimately to become the Royal Naval Scientific Service. He was the assistant and confidant of the first director of scientific research, Dr. (later Sir) Frank Smith, and of his successors. The last of these, Mr. (later Sir) Charles Wright, selected him in 1936 as his deputy, a post he filled for ten years. In 1946 he was appointed the first director of research programmes and planning, with special responsibility for the management of peace-time research and development expenditure. Successive controllers of the Navy have spoken warmly of the skill with which these duties were carried out. He served on various Admiralty committees and represented the Admiralty on outside committees, always with distinction. His valuable advice was sought officially and unofficially by high and low on almost any matter which was 'tricky', and what he did behind the scenes often determined the success of actions in which officially he took no part. His many important works were recognized in 1953 by the award of the C.B.

Dr. W. E. Pretty

With the retirement of Mr. Buckingham, the Department of Research Programmes and Planning has been absorbed into a new Department of Research and Development Services ; Dr. W. E. Pretty has been appointed to be its first director. As a Royal Scholar, Dr. Pretty was awarded the A.R.C.S. and the Governors' Prize in physics at the Imperial College of Science and Technology in 1925, and in the same year took his B.Sc. (first-class honours), London. For the next six years his time was spent between teaching and research at the College under Prof. A. Fowler. His researches in spectroscopy gained him his Ph.D. degree. In 1931 he joined the Science Museum as assistant keeper in charge of the optics and the astronomy collections. He served as secretary to the Advisory Council of the Museum and was much concerned with the successful introduction of a new type of 'special exhibition' showing really up-to-date science and technology in a particular field. In 1939 he was seconded to the Admiralty Department of Scientific Research and at the end of the War accepted an invitation for permanent transfer to the newly formed Royal Naval Scientific Service. He was promoted to the rank of deputy chief scientific officer and appointed director of research programmes and planning in 1948.

Toxic Chemicals in Agriculture

The Minister of Agriculture, Fisheries and Food, with the Minister for Science, the Secretary of State for Scotland and the Minister of Health, have appointed the following Research Study Group to consider this subject : Prof. H. G. Sanders, chief scientific adviser (agriculture), Ministry of Agriculture, Fisheries and Food (*chairman*) ; Dr. H. R. Barnell, chief scientific adviser (food), Ministry of

Agriculture, Fisheries and Food; Dr. J. M. Barnes, director, Toxicology Research Unit, Medical Research Council; Prof. A. R. Clapham, professor of botany, University of Sheffield; Dr. R. A. E. Galley, director, Tropical Products Institute, Colonial Office (officer-in-charge, Colonial Pesticides Research); Mr. C. O. Harvey, Laboratory of the Government Chemist (Department of Scientific and Industrial Research); Dr. Donald Hunter (physician to the London Hospital and part-time physician-in-charge, Department of Research in Industrial Medicine, London Hospital); Mr. W. C. Moore, director, Plant Pathology Laboratory, Ministry of Agriculture, Fisheries and Food; Prof. W. L. M. Perry, professor of materia medica, University of Edinburgh; Mr. G. G. Samuel, scientific staff (headquarters), Agricultural Research Council. The terms of reference for the Group are: "To study the need for further research into the effects of the use of toxic chemicals in agriculture and food storage, and to make recommendations". The Group is independent of the interdepartmental Advisory Committee on Poisonous Substances used in Agriculture and Food Storage, which will continue.

Corrosion Science Society

THE Corrosion Science Society has been founded with the following immediate aims: (1) to promote the advance of corrosion science and its application to the solution of practical problems; (2) to organize meetings—personal, local, national and international—of scientifically qualified persons interested in all fields of corrosion and corrosion-protection research and practice, for the discussion of current work; (3) to co-operate with all other professional individuals, societies and institutes, British and foreign, with interests in the corrosion field. The first meeting will be held during April 4–5 at the Battersea College of Technology, London. Stress-corrosion, cracking, the corrosion and protection of aluminium, cathodic protection, electrode processes and high-temperature corrosion and oxidation will be among subjects to be discussed on the basis of a series of talks by workers at present active in these fields, including C. Edeleanu, J. G. Hines, T. P. Hoar, J. E. O. Mayne, N. F. Mott, E. C. Potter and L. L. Shreir. Those interested should write to either Dr. T. P. Hoar, Department of Metallurgy, Pembroke Street, Cambridge, or Dr. L. L. Shreir, Battersea College of Technology, London, S.W.11.

Research in the German Federal Republic

ON behalf of the Federal Republic, the German Embassy in London publishes quarterly an attractive journal, *Germany*. A recent issue was devoted to fundamental research and its application (4, No. 12; 1959). The main articles deal with plastics and their place in the German economy together with accounts of the work being carried out in the different plastic research institutes. Prof. Curt Hoffmann, of the Institute of Oceanography, Kiel, is the author of an essay on marine production biology, and Prof. K. O. Kiepenheuer, of Freiburg, describes solar research in Germany. To inaugurate a series of portraits of German Nobel Prize winners, there is an illustrated account of the life and work of Prof. Otto Kahn. The journal also contains articles describing various aspects of cultural life in Germany, the most interesting portraying celebrations in connexion with the Schiller bicentenary in West Germany. The annual subscription to the journal is D.M. 20.

The Burma Research Society

DURING 1910 a small group of Burma residents, mostly Civil servants with varied interests as amateurs in the natural history, archæology, history and literature of the country, established a society to serve as a forum for their discussions and a *Journal* for the publication of their findings. The creation of the University of Rangoon ten years later brought many well-qualified recruits to the Society and the University gave it a permanent home. The thirty-one volumes of the *Journal* published between 1910 and 1941 include many original papers of first-class importance, but everything came to an end with the Japanese occupation. All records and stocks of publications went on the bonfire of the University Library and very few complete sets of the *Journal* seem to be in existence. The Society started up again in 1948, and it was a happy thought of its 700 members, headed by the president, the Hon. U Chit Thoung, Minister of Culture, to mark the fiftieth anniversary by holding a six-day conference during December 28, 1959–January 2, 1960. Twenty-one papers were read and a further fourteen contributed to be published in a single volume with the conference proceedings. Some fifteen overseas visitors were guests of the Burma Government and were afterwards taken to Pagan, ancient capital of Burma, incredibly rich in archæological and historical treasures, still in course of study by Prof. G. H. Luce and others. At the conference, Britain was represented by Dudley Stamp (professor of geology and geography in the University of Rangoon, 1923–26). To further mark the occasion, the Society is reprinting the more important papers of its pre-war *Journal*. There are to be three volumes—respectively History and Literature (published), Economic and Social, and Science, obtainable from the Society, c/o University Library, Rangoon, at kyats 20 (30*s*.) each. The Science volume includes eight geological papers, and a number on earthworms, tapeworms, algae and agricultural pests. A complete index to the *Journal* is also to be published.

Scientific Societies in the United States

THE U.S. Office of Science Information Service recently distributed a questionnaire to more than two hundred national scientific societies (*Scientific Information News*, 1, No. 4; Aug.–Sept. 1959). The questionnaire is part of a study designed to provide objective information about the human and financial resources of scientific societies and their publications. Data requested by the questionnaire include: (1) total membership and distribution of members among various membership categories; (2) titles, frequency, and subscription rates of society publications; (3) sources of income, including subscriptions and endowments. This study of scientific societies is the first step in a wide-ranging programme designed to provide a clearer picture of scientific communication in the United States. The results of two other National Science Foundation studies—one of the policies, practices and costs of professional scientific journals and the other of scientific man-power data gathered by the National Register of Scientific and Technical Personnel—will be correlated with the scientific society study to help the Office of Science Information Service carry out more effectively its task of fostering co-operation and co-ordination among scientific information sources.

Darwin's "Journal"

IN his "Autobiography", Darwin referred to a "little diary, which I have always kept", in which he recorded the work on which he was engaged, the time which he took in completing each of his researches, the books in which they were described, and his movements from place to place. The document itself was headed "Journal". Francis Darwin also referred to his father's "Diary or Pocket-Book" as having been of great help to him in writing and compiling "Life and Letters". He added that "it was unfortunately written with great brevity, the history of a year being compressed into a page or less, and contains little more than the dates of the principal events of his life, together with entries as to his work, and as to the duration of his more serious illnesses". Some important quotations have been published from the "Journal", such as the famous entry under the date 1837: "In July opened first notebook on 'transmutation of species'. Had been greatly struck from about month of previous March on character of South American fossils, and species on Galapagos Archipelago. These facts origin (especially latter) of all my views". Edited by Sir Gavin de Beer, the "Journal" has now been published as a Bulletin in the Historical Series of the British Museum (Natural History) (2; No. 1). Students of evolution will be grateful to Sir Charles Darwin and the Syndics of the Cambridge University Library for granting permission for this rich chronological story to be published.

Preserving the Amenities of Great Britain

AN interesting example of the zealous way the Council for the Preservation of Rural England safeguards the amenities of Britain is revealed in its report for 1958–59. Its Central Panel Committee has recently considered the implications arising from the fact that the output of pressed Oxford clay bricks is now approaching 50 per cent of the entire production of all bricks in England, Wales and Scotland. Enormous quantities of these bricks are in general use within an area at least as far as 160 miles distant from the main producing area in Bedfordshire. As the appearance of new buildings throughout this area is linked with the type of brick available, the Committee felt that the manufacture of a reasonably wide variety was a matter for which the industry had great responsibility. These views were expressed in a letter to the Pressed Brick Manufacturers' Association. In reply, the Committee was informed that several new varieties are now on the market, and it is hoped that this policy will be promoted by the Association. The recent production of an attractive grey facing brick by the London Brick Co., suitable for use in stone districts such as north Bedfordshire, and adjacent parts of Northamptonshire, is a useful addition to the varieties now available.

Nature Protection in U.S.S.R.

MR. G. ALIEV, of the Academy of Sciences of the Soviet Republic of Azerbaijan, has described the text of a new law for the protection of Nature which was passed in the Republic in June 1959 (International Union for Conservation of Nature and Natural Resources, Bull. 8; Oct. 1959). The Commission for the Protection of Nature of the Academy of Sciences played a leading part in preparing and supporting this law. The Act provides for the conservation of the resources of the inanimate world and of living species, as well as the constitution of nature reserves and partial reserves. The same Act specifically mentions some interdicts: prohibits all economic activity within the reserves which would be contrary to their objectives; all interference with the places and objects preserved; all sale and purchase of reserved territories; all breaches of the hunting and fishing regulations; all uncontrolled introduction of exotic species; as well as undertakings which might result in air or water pollution, or begin or aggravate the process of erosion, the last being a source of grave problems in Azerbaijan. Any violation of the Act is considered a breach of Article 138 of the national constitution, which aims at "protecting and strengthening the sacred and inviolable foundation of the Soviet regime: public and socialist property". The Council of Ministers of the Azerbaijan S.S.R. and certain other Government departments have been charged with a number of duties relating to the protection and restoration of natural resources as well as measures against pollution and erosion; scientific preparation of natural resources of development schemes, recommendations, as well as the spreading of the principles and aims of conservation of Nature.

Liquified Petroleum Gases

LIQUIFIED petroleum gases are petroleum hydrocarbons which are gaseous at normal temperatures and liquified by pressures of a few atmospheres; they are thus easily stored and transported as liquid in relatively light pressure containers. 'L.P.G.', as they are known, may be derived from wet natural gas containing propane, butane and higher hydrocarbons; refinery distillation, whence propane and butane are obtained from crude oil by pressure distillation; cracking and reforming refinery processes. In a paper by R. F. Horner, P. C. Jarvis and A. G. Turner, at a symposium on the Uses of Liquified Petroleum Gases at the Institute of Fuel during December 1959, are described composition and properties of 'L.P.G.'; historical market development; applications; supply and distribution; future uses in the United Kingdom. Among other advantages is facility of handling and burning; precautionary measures must be taken in design of plant, because of their higher vapour pressure than ordinary liquid fuels. Combustion characteristics of 'L.P.G.' are high calorific value, specific gravity and high volumetric proportion of air required for combustion compared with coal gas; low flame velocity and narrow limits of inflammability compared with coal gas, hydrogen or acetylene. In a complementary paper by J. P. Liotier at this symposium is an account of "Uses of 'L.P.G.' in Industry in France with Particular Reference to the Metallurgical and Ceramics Industries", following developments for commercializing propane in French industrial markets, originally sponsored by L'Utilisation Rationelle des Gax.

Progress in Microchemistry

THE *Microchemical Journal*, 3, 3 (August 1959), contains a collection of review articles on various aspects of microchemistry which might be considered to have a wider appeal than the normal quarterly volume of this journal. The collection, which is generally of a high standard, suffers from a great variation of treatment by the twelve authors who contribute the articles. As an example, the section dealing with "Progress in Elemental Quantitative

Organic Analysis 1958" is very much to the point and gives good coverage to the work in this very important field, while the article dealing with "Progress in Qualitative Inorganic Analysis 1958" is written as a piece of special pleading for the replacement of classical techniques by others recommended by the author without sufficient explanation for his choice. The bibliography covers a wide range and has more references to European work than is sometimes found in American publications. This is probably a reflexion of the world-wide interest in microchemistry. There appears to be no reference in any of the sections to the determination of traces of residual pesticides in animal and plant tissues. This subject, which has a literature as extensive as the whole of the volume under review, would have added to the general value of the collection and could have provided material for discussion of the determination of sub-microgram quantities of organic compounds. The short section on equipment refers exclusively to American instruments. Subjects covered are, microscopy, fractionation and inorganic, organic and biochemical analysis. On the whole this is a useful collection of papers.

Russian Review of Biology

The Institute of Biology, in collaboration with the Department of Scientific and Industrial Research, is to publish an English translation of the *Russian Review of Biology* (Oliver and Boyd, Ltd. £9 for nine numbers in all for 1959 and 1960, and £6 10*s*. per annum for six numbers thereafter). The Russian journal has been running for many years, and contains summaries, each dealing with progress in a specific topic of biological research. The first translated number starts with a polemical discussion of a work by Lenin in relation to Soviet materialist biology. This is followed by review articles on two enzymes (polynucleotide phosphorylase and xanthine oxidase), the use of enzymes in biological investigation, erythrocyte sedimentation and, finally, the immunological reaction in embryos and new-born animals. There are extensive bibliographies, which, incidentally, indicate the familiarity of Russian biologists with research throughout the world. This new translation will help to acquaint more English-speaking workers with the biological research going on in Russia. Under 'current events' there is an appreciative account of a visit by Kh. S. Koshtoyants to some of the biology departments at Cambridge.

Sarawak Museum

The *Sarawak Museum Journal*, Vol. 8, of June 1958 is edited by Mr. Tom Harrisson, Government ethnologist and curator of the Sarawak Museum, and includes some characteristic forthright editorial comments. The volume is divided into five parts—the past, the present, some recent Borneo writing reviewed, Borneo plants and animals, and climate. Although the majority of the articles are of interest chiefly to those who live in Borneo, others, such as a paper on pollen from the South China Sea by Dr. F. R. Van Veen, are of more general application.

Bronze Figures from Nigeria

An illustrated article on some bronze figures from Ita Yemoo, Ife, Nigeria, by Frank Willett, the archæologist of the Department of Antiquities, appeared in the November 1959 issue of *Man*. The finds were unearthed by chance, and luckily Bernard Fagg, the director of antiquities, happened to be in the vicinity. As a result, all particulars of their occurrence, etc., were collected, and the finds themselves carefully preserved. Like the pottery figures discovered some time earlier, they are of great interest, though, perhaps, not so beautiful. An interesting fact is that five of the bronze heads are gagged, and they probably represent victims for sacrifice in view of the sherds of a ritual pot found nearby. Doubtless all these various art discoveries in Nigeria will be published in book form in due course.

Mathematics and Children

An article by Z. P. Dienes, lecturer in mathematics in the University of Leicester, in *Educational Research* shows that there are ways in which mathematical concepts can be caused to develop in children so that the techniques they learn are preceded by an understanding of the corresponding mathematical structures (*Educat. Res.*, 2, No. 1; Nov. 1959). Dienes's scheme has not yet been tried out under actual school conditions, but it is possible to state some qualitative results derived from trials in a number of schools. Schemes of individual mathematical discovery can be worked in large unstreamed classes throughout junior schools; a large majority of normal children can understand the complex workings of the four rules of arithmetic in the decimal and other systems; normal 10-year-old children can learn the use of brackets, the properties of the square, the solution of linear and quadratic equations and the factorizing of quadratic functions, if approached constructively.

It is possible to give children leaving junior school a solid body of practical experiences, impossible to obtain from real-life situations, on which an extensive secondary school curriculum can be based. There are almost limitless possibilities for enlarging the junior school syllabus, by including such topics as theory of groups, usually reserved for university honours courses. There is sufficient evidence to suggest that mathematics can be introduced to a larger section of the child population than had hitherto been thought practicable or possible. This is done by co-ordinating the teacher's work with the natural process of concept formation in the child. It is a challenge to the teaching profession to devise ways in which this much larger body of children can be drawn into the mathematical workshop. By doing so, a large potential of future mathematicians, scientists, and technicians would be created, whose practical bias is likely to make them extremely useful members of an increasingly technological society.

Preventing Evaporation from Reservoirs

Vast quantities of water evaporate from storages located almost everywhere in Australia. Much of the pastoral land lies in regions where evaporation from a free water surface exceeds 6 ft. a year, and annual losses of 8 ft. are not uncommon inland. The Mansfield process, devised by Mr. W. W. Mansfield of the Division of Physical Chemistry of the Commonwealth Scientific and Industrial Research Organization (Australia), has provided pastoralists and farmers with a simple method of cutting these enormous losses by at least a quarter. The process involves spreading a very thin film of hexadeconal (a pure form of cetyl alcohol) over the surface of stock tanks and small farm dams. The film is continuously replenished as required from beads of hexadeconal housed in a small raft with gauze windows. Because of its enormous potential significance, details of the Mansfield process were released at an early stage in its development. This has allowed more experience to be accumulated under different conditions throughout Australia, and widespread use on thousands of dams has confirmed its potential value. However, further use has emphasized that good results are not obtained unless the raft used meets the Commonwealth Scientific and Industrial Research Organization requirements. The condition of the raft must be checked periodically. A recent article discusses some of these practical points, and describes how the few unforeseen difficulties that have cropped up can be overcome (*Rural Research in C.S.I.R.O.*, No. 28 ; June 1959).

Research in the Hat and Feltmaking Industries

The twelfth annual report of the Director of Research, British Hat and Allied Feltmakers Research Association, covering the year ended August 1959, stresses the addition to the research programme of a study of the shock-absorbing properties of felt and other materials, relative to protective headgear (Pp. 20. Droylsden, Manchester : British Hat and Allied Feltmakers Research Association, 1959). Other work, which has largely been in continuation of work already projected, has included further study of the carrotting of fur, an examination of the fibre properties of rabbit fur from different sources and attempts to assess the feltability of vials by simple tests that could be carried out in the factory. A theoretical study of the free oscillation of the beam system of the felt-hardening machine suggested that the pattern of behaviour when the weight is adjusted reflects the changes in the amplitude and natural frequency of vibration of the beam assembly. The experimental programme with selected polyethanoxy assistants in dyeing felt showed that the assistants were selective in action with acid-levelling, acid-milling and both 1 : 1 and 1 : 2 premetallized colours and even in a particular class. In studies of the relation between the size and shape of the hat body required to make hats of a given size, brim-width and crown-shape, wide differences were observed in the stretch pattern, and theoretical analysis indicated that the size and shape of hat bodies to give hats with a predetermined stretch pattern could be calculated if the geometry of the stretching could be stabilized. A review of the system of TDM indexes for measuring felt quality showed that the dependence of physical properties upon form weight changed with time and that periodic revision of the indexes was required to avoid obsolescence. A revised system has been proposed, while further work on frictional properties has provided an explanation of faults in felt which developed when bodies were pounced wet. Lists of publications, officers and staff are included.

"A Microsomal Malic Dehydrogenase"

Dr. Kenneth H. Shull writes : "It has come to my attention that three errors were made in my communication under this title in *Nature*, 183, 259–260, January 24, 1959. In both Figs. 1 and 2 the sentence in the legends referring to the abscissæ should read 'The abscissa is the reciprocal of the substrate (oxalacetate) concentration in $\mu M \times 1{,}000/3$'. The value of the Michaelis constant, *Km*, line 24, column 2, p. 259, should read $4 \cdot 66 \times 10^{-5}$ *M*. The oxalacetate concentration, line 9, p. 260, should read 253 μM. These errors arose from the fact that I plotted units of mass, micromoles of oxalacetate per cuvette, rather than units of concentration. Since each cuvette contained a total volume of 3·0 ml., these reciprocal values for concentration are too large by a value of 1,000/3. The Michaelis constant which was determined graphically from Fig. 1 is thus too small by this same factor".

Announcements

Sir Gavin de Beer will retire from the directorship of the British Museum (Natural History) on April 30. He will be succeeded by Dr. T. C. S. Morrison-Scott, at present director of the Science Museum, and previously on the staff of the British Museum (Natural History) in charge of the Mammal Room.

No. 11, Vol. 5 (November 1959) of "The Bibliography of Scientific Publications of South and South East Asia", compiled jointly by Unesco Science Co-operation Offices for the area and published by Modoc, National Physical Laboratory, New Delhi, contains a further 608 classified entries.

The second Asian/Pacific Congress of Cardiology will be held in Melbourne during the last week in May. Further information can be obtained from Dr. A. E. Doyle, Alfred Hospital, Melbourne, S.1, Victoria, Australia.

La Société de Chimie Physique is arranging a meeting on the Structure of Graphite and the Kinetics of its Reactions during May 31–June 3. Further information can be obtained from the Secrétaire Général de la Société de Chimie Physique, 10 Rue Vauquelin, Paris-V^{e}.

The series of Summer Schools in Theoretical Chemistry which are held annually at Oxford is being continued this year during September 10–24, under the direction of Prof. C. A. Coulson. Further particulars can be obtained from the Assistant Secretary, Delegacy for Extra Mural Studies, Rewley House, Wellington Square, Oxford. Applications should be made before April 30.

A symposium on Molecular Structure and Spectroscopy will be held at the Department of Physics and Astronomy, Ohio State University, during June 13–17. There will be discussions of the interpretation of molecular spectroscopic data as well as methods for obtaining such data. In addition, there will be sessions devoted to those phases of spectroscopy of current interest. Further information can be obtained from Prof. R. A. Oetjen, Department of Physics and Astronomy, Ohio State University, Columbus 10, Ohio.

A two-day conference, to be held in Philadelphia during April 28–29, on "The Role of Nuclear Propulsion in our Future Merchant Marine", has been arranged by the Atomic Industrial Forum, an association of the atomic industry in the United States, in co-operation with the U.S. Atomic Energy Commission and the Maritime Administration. The provisional agenda includes : economics of nuclear ships, safety and liability problems, progress in foreign nations, and future prospects for the U.S. programme. Further information can be obtained from the Conference Manager, Atomic Industrial Forum, 3 East 54th Street, New York 22.

SCIENCE IN CIVIL LIFE IN BRITAIN

TWO recent debates in Parliament are of special interest for the further light thrown on the Government's intentions with regard to the Minister for Science. The first of these, in the House of Commons on December 7, was the outcome of a supplementary estimate not exceeding £4,080 for the Office of the Minister for Science for 1959–60, moved by Sir Edward Boyle, Financial Secretary to the Treasury. Sir Edward explained that this addition to the original Vote of £36,090 covered the remaining cost for the staff of the Atomic Energy Office and the Lord President's Office, and in particular the new provision for a chief information officer and supporting staff and provision of additional staff for the former Lord President's Office. He pointed out that Lord Hailsham's responsibility for the Atomic Energy Authority is in a sense a reversion to an earlier practice; but his responsibility for the four great executive research councils involves a considerable volume of Government expenditure; for the Department of Scientific and Industrial Research, the current expenditure, he said, is £13·25 million; for the Medical Research Council £3·5 million and for the Agricultural Research Council, just under £6 million. The Atomic Energy Authority accounts for £55 million current and £37 million of capital expenditure. The Minister for Science is also responsible for the Advisory Council on Scientific Policy and its Scientific Man-power Committee, the Overseas Research Council, and the Steering Group for Space Research; and Sir Edward Boyle emphasized the close relation of the Advisory Council for Scientific Policy both with the National Research Development Corporation and with defence science, and said that Lord Hailsham has already asked the Council to advise him on the balance of our national scientific effort. Within the four main research councils there are 97 research stations and establishments, and Lord Hailsham attaches great importance to links between these establishments and the universities and technical colleges, and will surely make these links and relations as fruitful as possible.

So far as defence research is concerned, the care of such activities, especially in the field of aircraft, guided weapons and electronics, are to be transferred from the former Minister of Supply to the new Minister of Aviation; but the responsibilities of the Minister for Science for space research will ensure very close contact with this work. Sir Edward also anticipated that the Minister would use the establishments of the research councils to reinforce the work of numerous Ministries and give them general guidance and advice in matters of scientific policy; but he suggested that the greatest challenge of all is to ensure that modern ideas and discoveries in science and technology are effectively and rapidly applied in British industry, and in this he thought development contracts may prove of service.

In the debate, some doubts were expressed as to the adequacy of the scientific staff in the Minister's office, and Mr. A. Greenwood, while welcoming the tribute paid to the Advisory Council on Scientific Policy, referred to the failure to implement the Council's recommendation regarding a national scientific reference library and suggested that the Council might need a stronger secretariat and an intelligence unit of its own. He and almost every speaker in the debate was critical of the absence of a spokesman for the Minister in the House of Commons. Sir Arthur V. Harvey referred to the need for greater co-ordination, mentioning especially the Faery rotodyne and Jodrell Bank Experimental Station. Mr. A. Albu stressed the need for much more money for basic research and for carefully selecting the lines of development on which our resources should be concentrated. Mr. Robert Carr suggested that the Minister might look into the possibility of stimulating research in industry through research contracts, and was concerned as to the possibility of too great a split between defence science and civil science under Government control. He also stressed the need for an adequate supply of technicians and skilled workers, and hoped that before long one Minister, instead of the present six, would answer in the House of Commons for the Minister for Science. Both Mr. Facey-Jones and Mr. Chuter Ede referred to the importance of full publication, Mr. Ede pointing out that secrecy is the great enemy to the progress of science, and Mr. D. Price stressed the value of an understanding of science in the Civil Service.

The Minister of Education, Sir David Eccles, replying on the debate, was able to show good reason why he should reply for the Minister for Science, particularly in view of his responsibilities for the schools and technical colleges. He believes there is a good prospect of reaching our target of 20,000 scientists and technologists. Although we are still short of science teachers, the numbers in grammar and technical schools are improving, and in 1959–60, 865 graduates in science entered university departments of education compared with 639 two years ago. He thinks that the Minister for Science will collect and co-ordinate relevant views on the controversial issues of scientific man-power, and place his conclusions before the education departments, universities, other scientific bodies and industry, and that he would be in a better position than any other Minister or agency to form estimates of the scientific resources and needs of Great Britain. The appointment of a Minister for Science has inevitably created some awkward administrative problems, but with good will there is no reason why they should not be settled.

The second debate, on science in civil life, opened by Lord Taylor in the House of Lords on December 9, is of greater interest because it provoked a further account from Lord Hailsham himself of his conception of his post as Minister for Science. Lord Taylor, after pointing out that it was no new thing for a senior Minister to be in charge of the main civil scientific Departments, referred more particularly to medical science and expressed the opinion that the Medical Research Council is a model of Government intervention in the sciences. Nevertheless, he thought that too much paper work comes to the Council for final decisions, and that the Council sometimes fails to try out new ideas outside the conventional rut of thinking; he also suggested that besides, as a broad principle, backing good men, it might more often suggest the problems to be tackled. In this connexion, Lord Taylor named three specific projects and then referred to a third difficulty arising in relation to the universities, through the shortage of endowed readerships and research professorships, and the consequent loss of good research workers in the medical field to clinical medicine. Universities, he said, are also unwilling to recognize Government research units and organizations as approved places for study for higher degrees, and he mentioned specifically research hospitals or hospitals with a specialized research unit. Like several subsequent speakers, he urged the need for a great effort in the social sciences, and suggested the establishment of a research council for the social sciences.

Lord Hailsham welcomed Lord Taylor's suggestions as illustrating the immense range of opportunity, and, replying to a specific question about scientific attachés, said that a fifth scientific attaché would be appointed, this time in Moscow. Dealing with the question in its broadest aspects, he said that the relation between Government and science and technology in all its aspects is one of the great challenges to the present Parliament. He himself is anxious to be as accessible as possible to members of both Houses, and insisted that the creation of his post was an evolutionary rather than a revolutionary step, and that no one should be dogmatic about the direction in which this kind of organization would lead us. Of his Ministry, he observed that there are only 39 people concerned with the expenditure of some £112 million of public money, and the executive work is carried out by scientists with a relative freedom, though subject to Government direction of one kind or another.

Looking further at the nature of the challenge to which he referred, Lord Hailsham said that science is not something which can be disentangled and treated separately from life. The approach of a scientific society and the need to learn how to operate this society and understand what it means involves talking in terms of generations rather than parliaments. What we do and say about a scientific society must be set against the present background: perhaps twenty or thirty years of intensive education, training and effort have been required to produce the men upon whom ultimately this kind of progress in human knowledge will depend. Lord Hailsham pointed out that this small group of pioneers needs the support of much more scientific literacy in the governing elements of the country and accordingly of our whole society, and he stressed the importance of the educational system developing such literacy. Science also has to be sustained by an immense army of skilled craftsmen and technicians, and while the Government must be responsible for the whole and for finding and strengthening the weak places, this must also be the joint responsibility of all concerned with the administration of any part of our society. Above all, scientific work must and can only be administered by scientists. Lord Hailsham also said that the distinction between the various defence and civil sciences is wholly artificial and arbitrary, and that scientists would see in a Minister for Science a real assurance that the civilian applications of science would not be overlooked because of the greater expenditure on defence projects. Science and scientists are not solely concerned with national things, and the values of the scientist are not purely material. Replying to Lord Morrison, Lord Hailsham firmly vindicated the creation of his office as Minister for Science; but he thought the proposal for a social science research council might be premature. He agreed with Lord Adrian as to the importance of an adequate supply of technicians in order to ensure efficient use of the services of scientists and technologists, though this presents some difficulty at the universities, and he also agreed as to the importance of sciences that do not fall into the normal classification. The Advisory Council on Scientific Policy has already been asked, in looking at the general balance of the scientific effort in Britain, to pay special attention to oceanography, seismology and astronomy and the relative stress to be placed on biology. He also firmly maintained the soundness of the decision to limit our effort in space research and emphasized the importance of international co-operation in this field. He agreed with Lord James of Rusholme as to the importance of science teaching, and that although accommodation is less important than the provision of teachers, it could become a limiting factor and therefore requires attention. Lord Hailsham hoped that during the coming session in Parliament there will be some debate on the size of universities and their contribution to our scientific effort, and that the present debate would be only the first of many on science in civil life.

SCIENCE AND FOREIGN POLICY IN THE UNITED STATES

IN the years since the Second World War, non-military scientific developments have played important parts on the international stage. With technology accelerating at an amazing rate, what effects will it have on foreign relations problems in the future? Can we look ahead and try to ascertain what science will produce, and then take advantage of the product in foreign policy planning? These were questions posed by the Committee on Foreign Relations of the United States Senate (*Research for Industry*, 11, No. 6, Nov.–Dec., 1959). Stanford Research Institute scientists in all divisions provided assistance to Eugene Staley and Guy Benveniste in attempting to find some of the answers. The team indicated in its report that, while some future scientific developments may help solve foreign policy problems, the net effect of technological progress in the next decade is likely to be to create or intensify such problems. Thus, the security and well-being of the United States calls for a re-appraisal of present effort with the view of directing more energy toward non-military foreign policy challenges. Foreign policy planning should include continual review of scientific advances and their significance for international relations.

A specific suggestion was to establish a research and development office in the Federal Government oriented to identifying problems in newly developing countries and then stimulating research on these problems. In addition, an international development year might be organized along the lines of the International Geophysical Year with the aim of assisting newly developing nations on a grand, concerted scale. In order to utilize science and scientists to facilitate understanding between nations, a United Nations weather and climate agency might be established to carry on research and ultimately to have authority over all weather and climate control undertakings. A United Nations university or system of universities could be dedicated to the advancement of science and the humanities. Other suggestions envisaged are the creation of an international translation and scientific information centre, and the establishment of adequate facilities to conduct research on technical problems of disarmament.

This study is the second of fifteen the Senate Relations Committee has authorized to examine United States foreign policy. It is designed to provide the Committee with background information, and the members are free to accept or reject the findings. The chairman, J. W. Fulbright, has announced that public hearings will be held later "to receive testimony from all interested parties in order to test the soundness of the findings and recommendations in all the studies undertaken".

NUTRITION IN EUROPE

A SYMPOSIUM on "Education and Training in Nutrition in Europe", arranged by the Food and Agriculture Organization of the United Nations and the World Health Organization through its European Regional Office, was held in Bad Homburg, West Germany, during December 2–11, 1959. There were some sixty participants from twenty-two European countries, with Israel and Tunisia also represented. They belonged to a number of professional groups and included nutritionists, dietitians, physicians and public health officials, educationists, home economists and agronomists. The symposium was, in fact, designed to secure an inter-disciplinary approach.

The Food and Agriculture Organization and the World Health Organization are both concerned with nutrition ; the former because its basic objective is to satisfy nutritional needs by making the necessary foods available, the latter because of the relation of nutrition to health. For many years they have collaborated in programmes and projects in this field, under the guidance of a joint expert committee. The meeting in Bad Homburg was one of a long series of joint undertakings.

The symposium began by attempting to define the nature and extent of problems of nutrition in Europe, a task facilitated by reports prepared beforehand by the participating countries. Generalization was naturally found to be difficult, in view of the wide differences in the countries represented. For northern, western and central Europe the picture is, on the whole, a favourable one. Under-nutrition and severe malnutrition are rare nowadays, the temporary food shortages of the War have been replaced by abundance, perhaps even by over-abundance. The child of to-day is more wisely fed than his predecessor of thirty or forty years ago and a number of food-deficiency states and diseases once prevalent have been largely eliminated ; it was noted, for example, that in certain countries teachers of medicine have great trouble in finding cases of rickets to demonstrate to medical students. However, even in the best-fed countries the goal of food for health has not been fully attained. For example, dental decay was mentioned as a widespread and distressing condition due to dietary errors, though unfortunately there is not yet general agreement on what these errors are. Again, over-nutrition and obesity are in a sense a form of malnutrition, and there was agreement both on their increasing prevalence and danger to health. The still unresolved problem of the relation between fat intake and the modern scourge of coronary disease was regarded as being somewhat outside the terms of reference of the symposium, but its importance was stressed by many members. It was pointed out that the increasing proportion of elderly people in the population presents dietary problems to which insufficient attention has as yet been given.

In other parts of Europe malnutrition still occurs in various groups of the population and in certain areas within the countries. The groups most affected are infants, young children and expectant and nursing mothers. It was reported that rickets is common in Poland and that pellagra is still found in a few pockets in eastern Europe.

More information on existing food consumption and state of nutrition is required, to help Governments in establishing national food policies which take account of nutritional needs, and to guide efforts to improve diet through education. With regard to the latter, however, mere average data on the quantities of various foods which people eat, and their calorie and nutrient value, are not enough. It is also necessary to understand *why* they eat what they do in the way they do. Food patterns are influenced by tradition and by a host of economic, social and psychological factors of which the 'educator' must be aware. The symposium entertained itself from time to time by exchanging anecdotes on the extraordinary ideas which people often have about food and diet.

After analysis of the situation had indicated that there is plenty of room for improvement and a wide need for public enlightenment, the symposium considered the adequacy or otherwise of the training in nutrition now given to various professional groups, dealing *seriatim* with nutritionists, dietitians, medical students, public health personnel, home economists, school teachers and agronomists and agricultural extension workers. The nature and content of training were discussed under each heading, after an introductory paper presented by a chosen member. Professional nutritionists (of whom there are all too few) need knowledge which is deep enough to enable them to provide leadership and guidance in their countries. Dietitians, defined as a class of worker concerned with organizing the feeding of groups and individuals, in particular the sick, also need extensive specialized training. With respect to most of the other groups, the symposium, aware of the need to avoid the further over-loading of curricula, did not favour adding nutrition to the curriculum as a separate subject ; it expressed preference for teaching it as part of other related subjects, provided the knowledge so imparted is inter-linked and becomes an 'integrated' whole. On the training in nutrition now given to medical students, the views of those present at the symposium were unfavourable; it was noted that while students receive some instruction in the science of nutrition in its clinical aspects, this is not given in an 'integrated' way and is rarely related to the everyday problems of family life.

Emphasis was placed on the contribution to popular education which can be made by the public health nurse through her contacts with families in their homes. Home economists, a growing professional group in a number of European countries, can also influence families directly. As part of her work to improve family living conditions, the home economist is concerned with the planning of the family budget, the preparation of family meals, the domestic production, storage and preservation of food.

The training in nutrition of school teachers, so that they can teach its elements to their pupils, at an impressionable age, was discussed at some length. Ways of including nutrition in the teaching of subjects such as biology, health education and home economics were outlined, and it was agreed that it should not form a special and separate addition to school curricula. The need for simple and practical instruction was stressed, and attention was directed to the educational value of school meals.

Since good nutrition depends on the supply of enough of the right sort of food, it was appropriate for the symposium to consider the training in nutrition of agronomists and agricultural extension workers and their role in national nutrition programmes. Students in agricultural colleges learn much about the feeding of animals, but practically nothing about the feeding of human beings, which is after all the *raison d'être* of their profession.

Finally, the educational specialists present insisted that their colleagues should glance at modern educational techniques and modern ideas about the learning process, pointing out how these can be applied in the nutritional field. They were heard respectfully by a group in general more familiar with old-fashioned methods of instruction.

The symposium was aware that some of its conclusions cannot be exactly applied to the different countries represented, but require adaptation to the circumstances and needs of each. In the animated discussions, however, a large measure of agreement was reached on major points. Throughout, the friendly atmosphere characteristic of international technical meetings prevailed. W. R. AYKROYD

ELECTRON–PHONON INTERACTIONS

A CONFERENCE on Electron–Phonon Interactions in Solids was held at the Clarendon Laboratory, Oxford, during December 17–18. The Conference was held under auspices of the Physical Society and the delegates were welcomed by Dr. K. A. G. Mendelssohn (Oxford) on behalf of the President of the Society. The subject-matter of the Conference was of a very specialized nature and the organizers had further restricted the field of discussion by excluding topics directly related to superconductivity. It was therefore most encouraging that more than one hundred delegates attended the Conference. Among these were visitors from the United States, Belgium, Canada, Holland, South Africa and Switzerland. These delegates were not all specialists in the subject-matter of the conference, but nevertheless the discussions which followed the formal papers were often lively and to the point.

The Conference devoted its attention to those aspects of electron–phonon interactions which are most closely concerned with the complicated processes usually classified as 'transport phenomena' in metals and semiconductors. Examples of such phenomena are the electrical and thermal conductivities or the thermo-electric effects. The classical kinetic theory of the 'free electron gas' gives a very simple and elegant explanation of most of these effects in a first approximation. However, this theory makes the very fundamental assumption that the electron gas is in thermal equilibrium at some particular temperature, but does not clearly describe a mechanism by which this equilibrium can be achieved. The approach to equilibrium is discussed in terms of 'collisions' which do not involve interactions. The difficulties of a more rigorous theoretical treatment were emphasized by Dr. G. V. Chester (Birmingham) and Dr. J. M. Ziman (Cambridge). Dr. Chester, in attempting a more rigorous approach using the 'adiabatic' approximation, was rather surprised when experimentalists in his audience pressed him for some numerical orders of magnitude. Dr. Ziman reviewed the different methods at present used for estimating the electron–phonon interaction. He suggested that the strength of this interaction in metals was related to the Fermi energy; but the discussion which followed Dr. Ziman's paper showed that some delegates thought that he may have over-simplified the problem by laying such stress upon one parameter—the closeness of approach of the Fermi-surface to the Brillouin-zone boundary.

Prof. F. J. Blatt (Michigan State University), Dr. D. K. C. Macdonald (National Research Council, Ottawa), and Dr. J. R. Drabble (General Electric Co.) presented papers dealing with experimental investigations of thermo-electric effects in metals and semiconductors. Prof. Blatt described an interesting series of experiments which had been carried out using alloys of zinc, germanium, cadmium, indium, tin and antimony with copper. Of these alloys, 1 per cent zinc or germanium alloyed with copper gave a change in the thermo-electric power consistent with the expected phonon drag contribution estimated from pure copper. On the other hand, the addition of 1 per cent cadmium, indium, tin or antimony to copper reduced the phonon drag contribution almost to zero by virtue of phonon scattering arising from the solvent mass difference.

On the morning of the next day, Dr. A. B. Pippard (Cambridge) gave a simplified theoretical treatment of the anomalous absorption of ultrasonic waves by conduction electrons when the metal is situated in a magnetic field. Ultrasonic waves are essentially very long wave-length phonons, so that this technique provides in principle a direct approach to electron–phonon interaction processes. Unfortunately, this anomalous absorption is difficult to measure and is often masked by other attenuating mechanisms such as may arise from the movement of dislocations. The effect sought for has apparently only been observed in copper and possibly in lead. Other papers during the morning were contributed by Dr. R. Furth (London) and Dr. L. Mackinnon (Leeds).

The session during the afternoon was opened by Dr. S. Raimes (London), who gave a careful account of the description of transport phenomena in metals using the techniques of plasma theory. This was followed by a paper in which Dr. R. J. Elliott (Oxford) directed attention to an alternative line of investigation which might provide useful information about electron–phonon interactions. He suggested that further electron-spin resonance experiments should be undertaken with metals in order to measure the electron spin-lattice relaxation time in more detail. The final paper was given by Dr. D. M. S. Bagguley (Oxford). He described some experiments using cyclotron resonance techniques to investigate the different collision processes for carriers in germanium.

All the delegates who attended the conference were grateful to the organizers for their efforts in providing an opportunity for listening to the formal papers and for informal discussions between colleagues. Such informal discussions are themselves a sufficient and complete justification for a conference of this nature.

D. M. S. Bagguley

BRITISH GELATINE AND GLUE RESEARCH ASSOCIATION

THE eighteenth meeting of the Research Panel of the British Gelatine and Glue Research Association was held on November 12 with Mr. J. N. Blake (Richard Hodgson and Sons, Ltd.) in the chair. Mr. Blake introduced Dr. D. A. Sutton, who had recently succeeded Mr. A. G. Ward as director of research.

Dr. J. E. Eastoe (Department of Dental Science, Royal College of Surgeons of England) presented the first paper entitled "Bone—The Structure of a Biological Microcosm". After considering bone as one of the several mineralized biological tissues, note was taken of its occurrence in the vertebrate subphylum of the chordate phylum. Dr. Eastoe discussed bone at descending orders of size beginning with the skeleton, its function and the integration of its component parts, and continuing through the cellular level to electron microscope studies of bone mineral and collagen and thence on to the organization at the molecular level of these two main components. He concluded with a discussion of the hypotheses advanced to explain mineralization. Particular reference was made to the work of Glimcher, Hodge and Schmitt showing that mineralization is induced from metastable solutions containing calcium and phosphate only when the collagen fibrils present have the 640-Å. spacing, and that deposition can be prevented by traces of polysaccharide components. Chemical differences exist between bone and skin in that the former contains less polysaccharide and that its lysine and hydroxylysine have ε-amino groups more reactive towards fluorodinitrobenzene according to recent work by Solomons and Irving.

In the second paper, "Shrinkage of Cherries in Jelly", Miss M. J. Anthistle (Fruit and Vegetable Canning and Quick Freezing Research Association) demonstrated that the shrinkage which has been noted on storage can be virtually eliminated by adjustment of the surrounding gelatine gel (isoelectric *p*H, 4·7) to about *p*H 4·0; below this *p*H shrinkage occurred and above it a slight degree of swelling. The effect was not dependent on the sugar concentration. Miss Anthistle then considered the phenomenon in terms of the Donnan membrane equilibrium theory and deduced that shrinkage should be minimal at the isoelectric *p*H of the gelatine; in fact, it was minimal between *p*H 4·0 and 4·5. The *p*H of minimum shrinkage is thus somewhat lower than the isoelectric *p*H of the gel. In the discussion, it was suggested that the slight discrepancy might be due to an effect upon the equilibrium caused by the protein and pectin inside the fruit.

A short paper was then given by Mr. A. M. Kragh (British Gelatine and Glue Research Association) entitled "The Equilibrium Moisture Content of Plasticized Gelatine Gels". Glycerine and a sugar mixture (4 sucrose : 1 glucose) were used as plasticizers and relative humidities of between 35 and 93 per cent were studied. Temperature had a negligible effect upon the equilibrium moisture contents. Unless the relative humidity was very high, the equilibrium moisture content of the mixtures did not vary greatly over a wide range (0·5 : 1–4·0 : 1·0) of plasticizer : gelatine ratios.

D. A. Sutton

INTERLINGUA

THE free circulation of ideas and information is not less important for science than the circulation of the blood is for the body. This circulation is impeded by the publication of the results of scientific research in a multiplicity of languages. At least 50 per cent of the scientific literature of the world is published in languages that more than 50 per cent of the scientists of the world are unable to read. The number of published articles is growing rapidly; chemical literature, for example, has doubled itself in the past eight and a half years. There is also an increasing number of international meetings. There is thus not only the difficulty of reading published articles but also that of participating in discussions with research workers from other countries.

According to Forrest F. Cleveland, the scientist has two requirements. First, he has to scan the current literature to obtain ideas and information with respect to new developments in his field. Secondly, he must read in detail those articles that pertain to his own investigations. These requirements are largely frustrated by the language barrier (*American Scientist*, 47, No. 3, 1959). Scientific work would be facilitated if the results of scientific research were published in a single language that every scientist would be able to read at sight. Such a tool of intercommunication became available with the publication in 1951 of the "Interlingua–English Dictionary" and the "Interlingua Grammar" (Storm Publishers, 80 East Eleventh Street, New York 3). This was the culmination of twenty-seven years of research by the International Auxiliary Language Association in New York. Interlingua was developed by taking the words of vocabularies of the European languages (including English), standardizing or rationalizing them to remove regional irregularities and peculiarities, and constructing a simple grammar for use with the resultant vocabulary. An international language was obtained, which in a large measure is already in use and which can be read practically at sight by every educated person. The basic criterion for the acceptance of a word for the Interlingua vocabulary is that it must be present in corresponding forms and with corresponding meanings in at least three of the four language units: (1) Italian, (2) Spanish, and/or Portuguese, (3) French, and (4) English—with German and Russian as possible substitutes.

Impressed by the easy readability of Interlingua, Cleveland began, in May 1952, the publication in this language of a small periodical, *Spectroscopia Molecular*. It contains brief reports of new research results, summaries of important articles in the current literature, reports of spectroscopic meetings, reviews of new books, a calendar of future spectroscopic events, details about new instruments, and news about spectroscopists and recent developments. The experience of this small periodical provides a proof of the utility of Interlingua for intercommunication in the world of science.

Perhaps the greatest use of Interlingua to date has been in medicine. More than twenty medical journals publish extracts in Interlingua with all their original articles. In addition, Interlingua has been used for summaries in the programme of seven international medical congresses. *Science News Letter* publishes each month a summary in Interlingua of recent scientific developments.

To assist editors and authors who wish to study the possibility of increasing the extension of their work by the use of Interlingua, Science Service, Inc., has established an Interlingua Division. This is located at 80 East Eleventh Street, New York 3, and is under the direction of Dr. Alexander Gode.

RESEARCH AND DEVELOPMENT

CAN the effectiveness of research and development be measured ? More precisely—given a certain amount of research effort, in a given field, can the rate at which useful results will emerge be predicted ? These are some of the questions asked during a recent intensive study, "Basic Research in the Navy," carried out for the Naval Research Advisory Committee by Arthur D. Little, Inc. (Industrial Bulletin of Arthur D. Little, Inc., 369, Nov. 1959). In its report to the Secretary of the Navy, the Committee recommended further "research on research"; one approach is through a mathematical model describing the mechanisms of discovery and application.

The model describes a two-stage process; in the first stage, knowledge is discovered, and brought from the realm of the unknown into the known. In the second stage. combinations of the known facts are fitted together and applied. Formulation of the inventive process in this way emphasizes one important point: for every invention there is a key fact, or relation, or principle—the last to be discovered of all those necessary before the invention could be made. From this it follows that the date of discovery of this key fact is the earliest date upon which the invention could be made at all—although, of course, many inventions are not made until long after the discovery of the key fact.

There is a body of available knowledge, containing a certain number of key facts, corresponding to an equal number of possible inventions. If no new knowledge is added by research, these represent all the inventions which can be made. But the more research effort is applied, the more new facts come to light. It is also proportional to the size of the pool of (undiscovered) knowledge. The consequences of this ratio are interesting; the equation describing the rate of depletion of basic knowledge—the unknown subject—is an exponential decay curve like that used to describe the rate of decay of a radioactive substance. This is the model for the first stage of the research process.

The second stage concerns the application of discovered facts through invention. Here again the rate of progress is proportional to the amount of effort expended and to the size of the pool of knowledge—in this case, the number of discovered, but unapplied, facts. Yet a fact may be applied many times, in different combinations with other facts. It is not the unit of knowledge that is important—rather, it is the conclusions drawn from a number of units. To determine a rate for the second stage properly, one must invoke the techniques of modern algebra.

There is, also, the question of the time-lag between discovery and application; for the second stage, as well as the first, one can draw an analogy with the process of radioactive decay. The body of known but unapplied facts is considered, on this model, to have a well-defined half-life, but the expected time for any one fact to be applied may range from very short to extremely long. The expected half-life itself is a variable quantity which depends on the amount of effort expended on development work.

By putting these various factors together, for the second stage an S-shaped curve is obtained indicating first a slow rate of application, then a more rapid increase, and finally a plateau; curves of this type are familiar to economists for describing the development of a market or a new product. This is the model for the inventive process, and it is confirmed by observation.

Perhaps the most interesting feature of the model is that an optimum ratio for the effort to be applied to the two stages of discovery and application of knowledge can be derived. That is, one can in theory answer the question: For maximum productivity of a research programme, what percentage of the budget should be devoted to basic research, and what part to applied research ? The model suggests that to develop one large and complex field, basic research should not fall below about 20 per cent of the total research budget.

UNITED KINGDOM HOSPITAL LIBRARY SERVICES

A PILOT survey of hospital library services has been made by an independent committee, of which Mr. J. A. M. Ellison-Macartney was chairman, sponsored by the King Edward's Hospital Fund for London (Hospital Library Services: a Pilot Survey. Report of an Independent Committee sponsored by King Edward's Hospital Fund for London. Pp. 56. London: King Edward's Hospital Fund for London, 1959. 2*s.* 6*d.*). Its report, now published, directs attention to the apathy of some hospital authorities and officials and to the inadequacy of much accommodation, but the greatest weakness uncovered by the survey was the quality of the books provided by the smaller voluntary organizations. The Committee believes that ideally the public libraries should be the major source of supply of books and that this should become a recognized function and duty of all such libraries. Nevertheless, the survey also revealed the great strength which lies in the supply of voluntary librarians, and the Committee also believes that ideally the staffing of hospital libraries could be undertaken by voluntary organizations in conjunction with the local public libraries supplying the majority of the books.

In general, hospital library services require a new look, and the Committee recommends that hospital authorities at the highest level should regard it as an essential duty to see that an efficient service is provided in all hospitals (except where special circumstances render it undesirable) rather than leave the decision to be taken at a lower administrative level. Far greater positive interest in the library service should be taken at all levels of hospital authorities and staffs; it should no longer be left in the main to voluntary helpers with the minimum of support in interest, funds and equipment. All architects' plans for new hospitals or extensions to existing hospitals should in future include library accommodation conveniently sited, and certain standards for accommodation and equipment are recommended for consideration. In particular, it is emphasized that because most library services are likely to continue to be run by volunteers, particularly women of middle age, the siting of library accommodation, height of shelves, weight of trolleys, heating, lighting and ventilation are of paramount importance. The standard of books provided should be that of books provided to the general public by the best public libraries, and the Committee recommends that for a hospital of average size, annual expenditure on new books should be not less than 6*s.* per bed. All hospital library services should include the provision of books to hospital staffs, and the Committee suggests that valuable training courses run by St. John–Red Cross should be open to all hospital librarians. The question whether books are capable of carrying tuberculosis infection requires scientific investigation and an authoritative pronouncement. The Committee believes that with the existing variety of library services, full efficiency cannot be attained on a basis of less than £1 per bed per annum at present prices, or £3 per bed per annum if salaries, but not overheads, are included.

THE ATLANTIC EEL PROBLEM

RECENTLY Tucker[1] published his view on the origin of the European eels. He advocates the revolutionary theory that all the European eels are the offspring of American parent stock. His argument is presented under three headings, of which the first, dealing with the return of the silver eels towards their breeding places, is in my opinion the most important one. Therefore, special attention will be paid to this section of Tucker's article.

Tucker begins by laying stress on the fact that the European silver eel is already well advanced towards being a reproductive oceanic fish despite the long journey which supposedly lies before it, this in marked contrast with the American silver eel, which is relatively retarded. The conclusion is drawn that this is just the reverse of what one would anticipate, taking into consideration the distance it has to go.

The impossibility of providing exact information on the time required to become a reproductive fish renders this subject elusive of criticism and debate. Comparison with other species of fish is moreover of little value; one should in my opinion concentrate on the features and habits of the European silver eel itself.

For the same reason the next paragraph dealing with the comparison of external features of both species under consideration cannot lead to an irrefutable conclusion. The only reasonable conclusion one could draw from the differences mentioned is, in my opinion, that there is every reason for believing in the existence of two really distinct species, which is just the opposite of what Tucker tries to demonstrate.

According to Tucker, the longevity of silver eels in captivity is irrelevant. We know, however, that the silver eels, kept in the tanks of the eel merchants, are always on the move; when the water in the tanks is in motion, the silver eels are swimming in the upstream direction. Despite the rather unfavourable conditions in the tanks, the silver eels go on swimming for months and months in succession. It is certainly a remarkable fact that such eels are, even after a period of prolonged storage, fat enough to be attractive for the consumer, for they can easily be sold at a fair price. This demonstrates that silver eels are able to move over huge distances without degeneration of their tissues. It seems therefore that this matter cannot be considered as irrelevant to the subject under discussion.

Two other items in Tucker's article are open to doubt:

(1) If one assumes with Tucker that the migrating eels must use a very simple orientation mechanism

based on currents, temperatures, and/or salinity conditions, and next, considering the complexity of the actual situation, concludes that such a system would fail to guide the eels to their supposed spawning grounds, this reasoning cannot be used as evidence to demonstrate that the eels will not reach such spawning grounds at all. Such a reasoning would indeed be comparable to the following: We assume that migrating birds orientate themselves on wind-direction and wind-strength. Since such a system cannot possibly guide the birds towards their destination, we find ourselves forced to conclude that birds do not reach their destination at all. Since we all know that the birds do reach their destination the only reasonable conclusion is that our assumption was not sound. Unfortunately we do not yet know how eels do manage to orientate themselves; but this also applies to birds: we do know for sure that they are capable of a perfect orientation but fail to understand just how.

(2) Tucker supposes that all European silver eels use the same pattern of behaviour and concludes that they therefore cannot all of them reach their destination. This conclusion is used to support his theory that European eels do not reach the Atlantic spawning grounds at all. Tucker, however, designs a migration system for the American silver eels; he finds himself forced to conclude that the Mississippi eels do not fit in this system. Tucker states then: "the fate of these need not concern us". If so, why does the fate of the Black Sea eels or the Baltic eels need to concern the migration system for the European eels ? I fail to find any logic in such reasoning.

Tucker then proceeds by putting the following question: "The eels if they survived would be abundant. Why have they never been taken by research vessels in the open Atlantic or by the intensive fisheries along the Continental Slope ? Are agility and this or that depth of swimming adequate explanations ?" I can adduce the following evidence in reply to this question.

Despite the alleged rapid degeneration the silver eel is an agile and 'clever' fish, which is extremely difficult to catch. In the IJsselmeer, on which I have worked as fisheries biologist for fourteen years, focusing mainly on eel problems, the only practical way to catch silver eels is with stationary gear, that is, fyke nets. With eel trawls, silver eels cannot be caught, though such nets are very suited to the capture of younger stages. Using the newly improved IJsselmeer eel trawl—especially designed and suited for catching eels in huge quantities—in the area along the Afsluitdijk we did not catch one single silver eel, notwithstanding the fact that the area must have teemed at that time with silver eels according to the catches made with fyke nets in the immediate vicinity.

Now the IJsselmeer is a shallow body of water, where silver eels abound, and use was made of a small meshed net equipped with two non-returning funnels. These conditions contrast sharply with those in the open and much deeper seas and oceans where silver eels do not concentrate but are sparse and gradually disperse. Fishing is carried out here with dragged gear, which we already know is not capable of catching silver eels even if the eels do not swim at "this or that depth" but would move along the bottom, as Tucker obviously supposes, but of which we are not at all sure. Further, the sea trawls are not equipped with non-returning funnels and have a mesh size in the cod-end of about 7 cm., big enough to offer even the biggest eel the best chance for escape. No fisheries expert acquainted with these details could expect the catch of a single silver eel in open sea.

If on the contrary the European silver eels die on the Continental Shelf, as Tucker presumes, millions and millions of dying and dead silver eels would occur locally on the sea bottom year after year. Undoubtedly such eels incapable of escape would regularly be scooped up by the numerous trawls scraping the sea bottom. As this does not happen we are forced to conclude that silver eels do survive their trek over the Continental Shelf. It cannot even be supposed that they die in deeper waters in the vicinity of the Continental Shelf, for such deaths will not occur at a given spot or depth but will be scattered over quite a vast area, considering the distances to be covered and the range in the maturating process.

A further argument of Tucker is that there are several deeps in the Mediterranean and the Atlantic Ocean, which offer conditions apparently identical to those of the Sargasso Sea; yet eels do not spawn there. I would point out that we do not know anything of the conditions for reproductive activities required by the eels; moreover, Tucker's argument is of the same type as the following: Scandinavian chaffinches in reality observed to hibernate in Ireland will have to migrate over England (which indeed is what happens); but since these birds do not hibernate in England, which offers conditions apparently identical to those in Ireland, it is highly improbable that they will hibernate in Ireland.

Tucker's discussion of the conditions which the American silver eels meet has—as I have already stated—no relation to the achievements of the European silver eels.

Later in Tucker's article are some statements which are not backed up by any substantial data. This also applies to the 'evidence' which is supposed to show the 'debility' of the European silver eel which is 'obliged' to drift passively down stream with the currents. However inactive they are according to Tucker, they are without any doubt very capable of escaping a highly specialized eel-fishing gear dragged through the water with a speed of several miles per hour. Apart from this we must reckon with the possibility that silver eels will be dragged along by the current in the direction of the Sargasso Sea, for a fair current is running in the opposite direction under the Gulf Stream at a depth of some 2,800 m., showing velocities up to 17 cm./sec. (that is, about 15 km./day)[2-4]. I wonder whether Tucker has been aware of this fact.

Tucker discusses some consequences—with rather far-fetched explanations—of the supposed non-return of the silver eels to their spawning places. In this respect I might direct attention to the necessity of constant hydrographical features, especially water temperatures, in the Sargasso Sea from year to year and that during scores of centuries (cf. also ref. 5). It is difficult to believe in such a feature, for the rather short period for which records are available allows us already to conclude that hydrographical conditions are not constant at all but fluctuate quite considerably in the oceans. Therefore one cannot assume with Tucker that a constancy of water temperature has been the guiding factor in the eel's reproduction.

Finally I would direct attention to Tucker's statement that: "The proposition of Schmidt[6]

(p. 204, lines 20–40: How do masses of larvæ in the Western Atlantic sort themselves out . . . . ?) has led to misunderstanding especially among lay authors".

It is quite incomprehensible why such a simple question should lead to misunderstanding, but anyhow it would have been fairer also to quote Schmidt's own answer: "In the main, the question is no longer difficult to answer. In the case of the American eel, the pelagic larval stage is terminated in about one year; . . . . It is otherwise with the European eel, which takes nearly three times as long over its larval development, as a result of which practically all of them are far away from the Western (American) portion of the Atlantic when the time comes for them, as elvers, to seek the coasts".

Surely, there can be no difficulty in accepting the fact that only fully grown leptocephali after having arrived on the slope of a Continental Shelf will stay there. After having completed metamorphosis they will continue their migration as elvers. The sorting out of larvæ off America is in my opinion quite plausible: the fully grown larvæ, that is, those of the American eel, respond to continental conditions and stay; all the others, that is, those of the European eel, with still two years growth ahead, do not respond, and hence are carried on by the Gulf Stream.

I believe that the meaning of all this is quite evident: the edges of the Continental Shelf constitute the area where tidal streams begin. Simultaneous with metamorphosis into the elver stage, the animals change their behaviour in such a way that it enables them to respond to, and take advantage of, the possibilities which the tidal currents offer them, as Creutzberg[7] has recently discovered.

For the sake of completeness I wish to remark that this is not the only known case of eels staying for some time in a transition area, next changing their behaviour pattern in such a way that they will respond to the newly encountered conditions and then continuing their migration. I discovered myself[8] that this is what happens when elvers migrate from sea into fresh water. The well-known metamorphosis from eel into silver eel is also a clear indication that the eel is a real master in the opportunistic art of changing habits and taking advantage of new conditions.

It will be clear from the foregoing discussion that Tucker's claim that his hypothesis is at parity with Schmidt's cannot possibly be supported by me. I still feel that Schmidt is right in his opinion. Concerning the still rather mysterious orientation of silver eels towards the Sargasso Sea, I am inclined to assume that silver eels, having a tendency to develop into a deep sea fish[9–10], are provided with an orientation mechanism directed westward. After leaving the European Continental waters in a westerly direction, many of the silver eels will inevitably meet the deep Atlantic current under the Gulf Stream but in opposite direction. When this happens I do not see any reason why the silver eels should not make use of this current, which will guide them afterwards towards the Sargasso Sea.

Such a journey might be of some 7,500 km. length. Supposing that silver eels will swim at a low speed, for example, some 50 km./day, of which they undoubtedly are capable[11], it will take them only about 150 days to reach their destination. We may be sure that such an effort exerted during that rather short period will be far from demanding the utmost of the animals and will undoubtedly leave enough reserve material to allow an ample development of the gonads.

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[1] Tucker, D. W., *Nature*, 183, 495 (1959).
[2] Deacon, G. E. R., *Oceanus*, 5 (3/4), 13 (1957).
[3] Veronis, G., *Oceanus*, 5 (3/4), 19 (1957).
[4] Swallow, J. C., and Worthington, L. V., *Nature*, 179, 1183 (1957).
[5] d'Ancona, U., *Nature*, 183, 1405 (1959).
[6] Schmidt, J., *Phil. Trans. Roy. Soc.*, B, 211, 179 (1922).
[7] Creutzberg, F., *Nature*, 181, 857 (1958).
[8] Deelder, C. L., *J. Cons. Int. Exp. Mer*, 24, 135 (1958).
[9] Svärdson, G., *Rep. Inst. Freshwater Res. Drottning*, 29, 129 (1949).
[10] Deelder, C. L., *Viss. Nieuws.*, 8, 43 (1955).
[11] Boddeke, R., *et al.*, *Proc. Kon. Ned. Akad. Wetensch.*, C, 62 (5) (1959).

Dr. Deelder is incorrect in referring to the American silver eel. This phase has not yet been observed, and the migratory stage is known as the bronze or black eel[1,2]. I understand from Dr. W. E. Frost that Dr. V. D. Vladykov recently identified the bronze phase as an intermediate stage between the yellow and silver in British eels.

The apparent maintenance of condition in 'stored' silver eels has already been discussed[3], together with the check on maturation imposed by artificially enforced life in fresh water[1]. 'Putte', the eighty-eight year-old Swedish eel, was in only partial nuptial dress when it died[4]. But under these conditions reserve fat, normally used for gonad maturation, locomotion and physiological (osmoregulatory) work[3], is available for swimming only and restricted swimming at that. It is not sufficient to observe vaguely that stored eels remain active and healthy and then claim that this "demonstrates irrefutably that silver eels are able to move over huge distances". We need concrete data on distances swum, speeds, weight-losses and analyses, preferably in salt not fresh water, and in the meantime it is more material that eels make the Baltic run at average speeds of between 5 and 10 miles per day and lose up to 20 per cent of their weight on the way[3,5].

The migrating bird argument is logical in itself but fallaciously applied. It is possible *a priori* to postulate and experimentally to demonstrate alternative methods of navigation in birds, and birds are known to arrive. It is possible *a priori* to reject all visual navigational aids in eels and, from our present knowledge of physics, oceanography and physiology, it is not possible to postulate any system other than one involving passive transport and inherent reactions to temperature and salinity gradients. Such a system could easily direct a majority of the American eels[1], but not the European: and European eels are not known to arrive.

"The fate of the Mississippi eels need not concern us"[1]. I was not forced to any conclusion whatsoever. Having shown a simple return route for the bulk of the American population I judged it immaterial and prodigal of space to deal withthe complicated Gulf of Mexico current system. Those who wish may study Dietrich[6] and evacuate their Mississippi eels through the Florida Current or through the more circuitous bottom currents of the Gulf and Caribbean. It is sufficient that we have put the major part of the American eels on to the spawning ground; we cannot devise any system that will put any section of the European population on to theirs.

Dr. Deelder's point about the IJsselmeer silver eel fisheries is well taken.

There is no real difficulty about the concealed mortality of silver eels in the sea, any more than there is with the normal mortality of herring, mackerel and all the other marine fishes. The sea has her predators to remove her sick, and her scavengers to keep her clean.

Since my comments on the fitness of the silver eel for migration have caused so much misunderstanding, I will now restate them.

The word 'debility' has been objected to by many other critics[3] besides Dr. Deelder, always through misinterpretation of its single appearance. The original text[1] reads: ". . . the debility of the (European) eel which is obliged to drift passively downstream in the current is due to demineralization . . . remineralization and resumed liveliness occur when the eel reaches the sea". Dr. Deelder chides me for lack of supporting data; I cited Bertin[7] (intending him to be read), who in turn gives a summary and bibliography of a dozen relevant papers by Fontaine and his school; both these authors also employ the word 'débilité' in the same context as I do, in relation to the late freshwater condition. There is no need for Dr. Deelder to attack with his IJsselmeer story; and is not the IJsselmeer still highly brackish ?

The adequacy of the stored food-reserves to provide for the journey is open to speculation[1,3,8]. The vitality of silver eels in inshore waters, re-affirmed by D'Ancona[8], Jones[3] and now Dr. Deelder, is immaterial to the problem of the ocean crossing. We are not interested in the condition of local populations inshore. We want to know the state of the residual European population by the time it crosses the Continental Shelf and is well on its journey, and we have no direct evidence as to that. But I maintain that the cases already cited[1,3,8], of eels losing weight on the Baltic run; of near-ripe eels, eels with degenerating guts, an eel with a blocked vent, provide material evidence that Baltic eels are too far advanced when only partly on their way. The same argument applies to precocious individuals taken from the Mediterranean. I argue that the condition of these silver eels, at most a few hundred miles from their home streams, is a significant indication of the probable state of the survivors of the whole continental European population when an equivalent distance out to sea.

I have already discussed *Conger conger* as a biological indicator of identical spawning conditions in various deeps in the western and eastern Atlantic and Mediterranean[3]. Dr. Deelder's chaffinch argument is logically and biologically unsound, as well as inapplicable to the present situation, but it is nevertheless worth analysing. The British Isles have only the native *Fringilla coelebs gengleri*, but are invaded each autumn by the related Continental *F.c.coelebs* as a winter migrant; the reverse migration of *F.c.gengleri* does not occur. Competing for the identical ecological niche each sub-species must clearly have some advantage over the other in its normal range. Such competition with members of both sub-species compels *F.c.coelebs* flocks to move on across Britain and into Ireland, where a seasonal increase is noted[9]. Now, there can be no competition for food or territory among spawning eels; there are no breeding populations of *Anguilla* in the eastern deeps; why, therefore should *A. anguilla* make a plainly hazardous migration to an area in which its larvæ must enter into competition with those of at least seven other Apodal fishes ?[2,10]. I think it is destroyed by its own metabolic failure before it reaches any of the likely spawning deeps, near or far.

I am mystified by Dr. Deelder's expressed doubt regarding my knowledge of the counter-current beneath the Gulf Stream; I did, in fact, specify this very current as the likely vehicle for the returning American eels[1]. The said current, running as it does near the foot of the American Continental Shelf, is irrelevant to the European eel problem unless Dr. Deelder postulates a return route via the Norwegian Coastal Current, Jan Mayen, and the Greenland Current, and so along the American eel route, with a greatly increased distance/time factor and a northward movement rather than a westerly one as he now specifically suggests. It is interesting to note here that tagged eels released in the Elbe have recently been recovered off Denmark, indicating a northerly rather than a westerly migration[11]. Dr. Deelder's estimate of likely 'low' migration speeds is, like Jones's[3], much too high in relation to those observed[3,6].

I am aware of the annual fluctuations in hydrographical conditions, but do not think that the broad Atlantic picture has altered sufficiently in (geologically) recent times to affect the validity of the new eel hypothesis. As regards earlier epochs there are further intriguing problems in the origin of the Japanese *A. japonica* population (which is closely related to[12] and may have been derived from the Atlantic *Anguilla* during the last Central American subsidence, or during an opening of the North-West Passage), and in the relation between climate and vertebral numbers of the European fossil *Anguilla*.

There is no need to labour my comment on Schmidt's account of the 'sorting' of the two types of leptocephali. In my general preparatory reading I found that it had been misunderstood and given rise to erroneous reasoning, and attempted a re-statement for that reason. There was no intentional unfairness in not quoting the passage in full to save space; again, the reference was intended to be consulted. Here I would like to correct an impression in certain quarters that my new interpretation of Schmidt's work is in some way derogatory to his memory. Dr. Anton Fr. Bruun, *in litt.*, has generously exonerated me of any such charge. If it were not for the oceanic researches of Dr. Johannes Schmidt and the experimental researches of the late Dr. A. Vedel Tåning, together with their other Danish colleagues, I would not have been able to write a word on this subject. Furthermore, most of the work which has been used in this re-assessment of the Atlantic eel problem was not published during Schmidt's lifetime.

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British Museum (Natural History),
London, S.W.7.

[1] Tucker, D. W., *Nature*, 183, 495 (1959).
[2] Vladykov, V. D., "Poissons du Québec", No. 6 (Quebec, 1956).
[3] Jones, J. W., and Tucker, D. W., *Nature*, 184, 1281 (1959).
[4] Lekholm, C. G., Halsingborgs Mus. årsbok, 163 (1939).
[5] Trybom, F., and Schneider, G., *Rapp. Cons. Explor. Mer*, 9, 51 (1908).
[6] Dietrich, G., *Z. Ges. Erd. Berlin.* 108 (1939).
[7] Bertin, L., "Eels: a Biological Study" (London, 1956).
[8] D'Ancona, U., and Tucker, D. W., *Nature*, 183, 1405 (1959).
[9] Witherby, H. F., Jourdain, F. C. R., Ticehurst, N. F., and Tucker, B. W., "The Handbook of British Birds", 1 (London, 1943).
[10] Bruun, A. F., "*Dana*" *Rep.*, 3, No. 9 (1939).
[11] Luhmann, M., and Mann, H., *Arch. fur Fischereiwiss*, 9, 200 (1959).
[12] Ege, V., "*Dana*" *Rep.*, 3, No. 16 (1939).

PROTECTION AGAINST ACUTE EFFECTS OF RADIATION BY HYPOXIA

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IT is well known[1] that the sensitivity of biological systems to radiation is increased in the presence of oxygen and decreased in its absence. In the case of whole-body irradiation of mammals, Dowdy *et al.*[2] have shown that exposure under reduced oxygen pressure results in a considerable increase of the *LD*50. In the course of a systematic study on the age factor in short-term and long-term effects of radiation[3], it occurred to us that the variation of radiation sensitivity with age, recently reported by us[4], may be associated with an oxygen effect. To test this hypothesis we investigated the survival of mice exposed to acute whole-body doses of radiation under hypoxic conditions. These measurements were carried out during April and May 1959, as a pilot experiment for a more detailed study of this effect in animals of various ages. Since, however, the completion of this investigation will take some time, it may be of interest to report our interim findings.

The mice used were of the same strain (albino *SAS*/4) as in the main experiment[3]. They were exposed in 'Perspex' containers consisting of twin cylinders, each of which could accommodate two mice. By means of tubes leading to a manifold, it was possible, by remote control, to substitute nitrogen for air in one of the cylinders. In this way four mice could be exposed simultaneously, two breathing air or oxygen and the other two nitrogen. To prevent convulsion of the mice while removing the oxygen, the mice were anæsthetized prior to irradiation. The anæsthetic used was pentobarbitone sodium ('Nembutal', Abbott Laboratories, Ltd.), at a dosage of 40 mgm. per kgm., administered intraperitoneally. No anæsthetic was used in the case of mice 1–3 days old, since they were not subject to convulsions under nitrogen. With this procedure we observed no ill effects of either the anæsthetic or the temporary hypoxia. Out of a total of 150 control mice which passed through the procedure of being anæsthetized and/or hypoxia, but without being given a dose of radiation, only two died in the course of thirty days.

The radiation used was the beam of 14-MeV. electrons from our linear accelerator. Electrons rather than X-rays were used to make it possible to deliver the required dose of radiation in a very short time. The mean dose-rate was about 2,500 rads per sec., so that even the highest dose employed could be delivered in less than a second. A broad electron beam was obtained by using an aluminium scatterer near the exit window of the accelerator. At a distance of 1 m., where the mice were placed, the beam was found to be uniform over the whole irradiated area.

For measurement of dose we used a small ionization chamber placed near the mice container. The dose absorbed was determined in two ways: (*a*) by a comparison of the readings of the ionization chamber with a 'Perspex' strip dosemeter which had been calibrated absolutely by a calorimetric technique[5], and (*b*) by calculating the collection efficiency of the ionization chamber and then making a correction for the conversion of e.s.u./c.c. into rads. These methods gave results in very good agreement with each other.

The experimental procedure, as determined by preliminary trials on the reaction of the mice to hypoxia, was as follows. After the container with the mice was placed into position for irradiation, the oxygen content was gradually reduced by admixture with nitrogen. This proceeded for 25 sec., by which time the gas was 100 per cent nitrogen flowing at a rate of 6 l. per min. The mice were kept in this atmosphere for 12 sec.; at the end of this time the exposure was made and the nitrogen immediately replaced by oxygen or air.

Figs. 1 and 2 show the results obtained with mice aged 1–3 days and 4 weeks, respectively. The percentage of the animals dying within 30 days is plotted on probability paper against the dose to which they were exposed. These graphs illustrate the striking protective effect of temporary removal of oxygen. From these and other graphs, the *LD*50 values for the different experiments were determined and are given in Table 1. For the tests with older animals

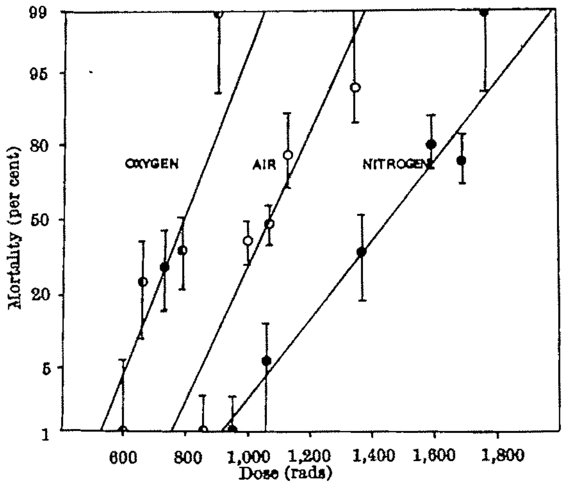

Fig. 1. Percentage 30-day mortality of mice 1–3 days old, breathing different gases, as a function of dose

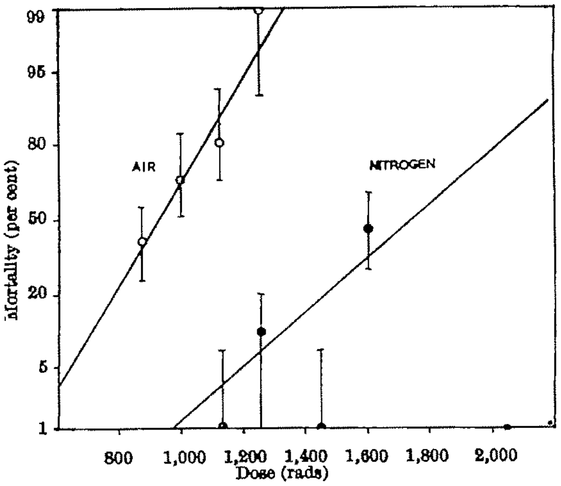

Fig. 2. Percentage 30-day mortality of mice 4 weeks old, breathing different gases, as a function of dose

Table 1. *LD*50 VALUES FOR MICE EXPOSED UNDER DIFFERENT CONDITIONS

Age	X-ray exposure in air (rads)	Electron exposure in :			Nitrogen protection factor
		Oxygen (rads)	Air (rads)	Nitrogen (rads)	
1–3 days	833 ± 24	788 ± 18	1,068 ± 20	1,450 ± 40	1·74 ± 0 07
4 weeks	684 ± 16	—	930 ± 45	1,760 ± 90	2·58 ± 0·15
19 weeks	880 ± 25	—	1,005 ± 65	1,750 ± 150	1 99 ± 0·18
82 weeks	764 ± 20	—	950 ± 100	1,320 ± 120	1 73 ± 0·16

very few mice were available; hence the large experimental errors for these groups.

Table 1 also contains the *LD*50 values obtained previously[4] for mice exposed under the standard conditions of our investigation, namely, using 15-MeV. X-rays at a rate of 7 rads per sec. (The conversion factor used (1 r. = 0·98 rad) was obtained in a direct measurement under the conditions of the experiment.) The last column of Table 1 contains the nitrogen protection factor, that is, the ratio of the *LD*50 at exposure in nitrogen to that under standard conditions.

These results show that exposure of mice under hypoxic conditions produces an entirely different age variation from exposure in air. For example, at 4 weeks of age the *LD*50 reaches a minimum when the mice are exposed in air, but it appears to have its highest value at the nitrogen exposure.

It is not yet clear how the oxygen effect operates in this case, but it may arise from differences in oxygen tension in tissues of animals of different ages. The relatively high *LD*50 value during the first week of life may be the result of a reduced oxygen tension in the neonatal stage, due to incomplete pulmonary ventilation and to circulatory readjustments of early extra-uterine life[6]. Indirect evidence of a lower oxygen tension is provided in our experiment in which the neonatal mice were exposed in an oxygen atmosphere; this gave a considerable decrease of the *LD*50. At normal oxygen tension the change should have been very small.

The increase of the *LD*50 for mice older than 4 weeks could also be due to a gradual decrease with age of the oxygen tension in tissues, starting from about 4 weeks. Such a decrease might result from changes with age in vascularity, diffusion coefficients and pulmonary ventilation. It is known that large gradients of oxygen tension exist in tissues, but there is little evidence that they change with age; there is also no evidence to the contrary. Another possible explanation of the results obtained with the hypoxic mice is that the time required to reduce to a given level the oxygen tension in the vital tissues, such as bone marrow, may vary with age, becoming longer the older the animal. This would mean that in our experiments the older mice were exposed under less hypoxic conditions than the younger ones. Experiments are now in progress to measure directly the oxygen tension in various tissues in mice of different ages.

Referring to Table 1, it will be noticed that the *LD*50 values obtained in air in the present experiment are, in all cases, higher than those obtained previously. The main differences between this and the previous experiment are: (*a*) electrons were used instead of X-rays; (*b*) the dose-rate was much higher; (*c*) the animals were, in some cases, anæsthetized. It is difficult to see how the change from X-rays to electrons could make a significant difference, particularly since the dose absorbed within the body of the animal was uniform within a few per cent. In other experiments, too, we have found no difference in the effects produced by X-rays or electrons at comparable dose-rates.

There remain the two other factors. We have evidence that anæsthesia does reduce sensitivity to radiation, probably because of a lowering of the oxygen tension in tissues[7]. This may account for some of the increase in the *LD*50 in animals exposed in air.

With regard to dose-rate it is known that over a large range of dose-rates the sensitivity to radiation increases with increasing dose-rate[8,9]. Our own preliminary investigations at high dose-rates do not show an increase of sensitivity; there are, in fact, indications of a decreasing sensitivity with dose-rate. Such a decrease, if confirmed, might occur for the following reason. Oxygen in the tissues is consumed during irradiation, but is normally replenished from the oxygen in the blood. If, however, the dose is delivered at a very high rate, the loss due to the first fraction of the dose may not be made up quickly enough and the remaining dose is given to the animal while some sensitive parts of it are under hypoxic conditions. Dewey and Boag[10] have reported such an effect in bacteria; the results of Kirby-Smith and Dolphin[11], who studied the chromosome breakage of *Tradescantia*, may also be due to a similar effect. If the same applied to mammals, then both factors, the anæsthetic and the high dose-rate, would really be oxygen effects. It is quite likely that the dose-rate effect would become manifest only in combination with an anæsthetic or a naturally low oxygen tension in tissues.

Further experiments, particularly using animals of more age-groups, are needed to elucidate these problems. Even from these preliminary results, however, it is obvious that age is an important factor in the protective action of hypoxia as well as in the acute effects of radiation. The same may apply to long-term effects. We have indeed evidence (to be published shortly) that long-term effects of radiation, such as shortening of life, show a similar variation with age at irradiation.

We wish to thank Mr. A. L. Bradshaw and Mr. G. S Innes for their help with the dosimetry and Miss Jeanne Moody and Mr. J. L. Bounden for their constant help with the experiment. We are also indebted to the Nuffield Foundation, the British Empire Cancer Campaign, and the Endowment Fund of St. Bartholomew's Hospital for grants which have made this work possible.

[1] cf Gray, L. H., "Lectures on the Scientific Basis of Medicine", 7, 314 (1957–58).

[2] Dowdy, A. H., Bennett, L. R., and Chastain, S. M., *Radiology*, **55**, 879 (1950).

[3] Lindop, P. J., and Rotblat, J., "Progress in Nuclear Energy", 47 (Pergamon Press, 1959).

[4] Crosfill, M. L., Lindop, P. J., and Rotblat, J., *Nature*, **183**, 1729 (1959).

[5] Boag, J. W., Dolphin, G. W., and Rotblat, J., *Radiation Research*, **9**, 589 (1958).

[6] Barcroft, J., "Researches on Pre-natal Life", 194 (Blackwell, 1946).

[7] Mack, H. P., and Figge, F. H. J., *Science*, **115**, 547 (1952).

[8] Corp, M. J., and Neal, F. E., *Int. J. Rad. Biol.*, **3**, 256 (1959).

[9] Lawrey, J. M., and Fowler, J. F., *Brit. J. Rad.*, **32**, 630 (1959).

[10] Dewey, D. L., and Boag, J. W., *Nature*, **183**, 1450 (1959).

[11] Kirby-Smith, J. S., and Dolphin, G. W., *Nature*, **182**, 270 (1958).

INTERPRETATION OF POTENTIOMETRIC TITRATION OF H-MONTMORILLONITE

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It has been observed[1] that the pH titration curves of hydrogen clays with strong monovalent bases have various characteristics, such as variation in steepness of slope in different parts of the curve. Variations have also been noticed for the kind of clay mineral, the amount of mineral in suspension, and the cations used to produce neutralization. Garrels and Christ[2], using original data of Marshall and Bergman[3] for the titration of the clay mineral (beidellite) in the Putnam silt loam soil, have shown that the neutralization curve of pH plotted against concentration of potassium hydroxide appears similar to that of two independent clay acids. They termed the two substrates the C^- (centre or interlayer site) and E^- (edge site). Blackmon[4] calculated exchange constants for the C^- and E^- sites of beidellite and 'illite' for K^+, NH_4^+ and Na^+. Cations are more strongly held by the E^- than by the C^- sites.

Neutralization curves obtained with different clay minerals show variations that can, in part, be attributed to the ratio of C^- to E^- sites. Differences in the bonding energy of monovalent and divalent cations introduce another variable that is expressed by the shapes of the titration curves. The resemblance between neutralization curves of some clay minerals and of humic acid suggested that a similar explanation would apply to these two kinds of substances, that is, the neutralization of a polymeric electrolyte.

Katchalsky and Spitnik[5] showed that the neutralization of a polymeric electrolyte could not be described by the Henderson–Hasselbalch equation:

$$pH = pK_a + \log \frac{\alpha}{1 - \alpha}$$

where pK_a is the negative logarithm of the ionization constant of a weak acid, and α is the degree of neutralization. Katchalsky and Spitnik ascribe this deviation to the uncoiling of the polymer chains during titration because weak hydrogen bonds are replaced by strong cationic bonds as ionization proceeds. These authors have further shown that a generalized Henderson–Hasselbalch equation of the form:

$$pH = pK_a + n \log \frac{\alpha}{1 - \alpha}$$

where n is a constant, will describe the titration of a polymeric electrolyte. It was therefore of interest to determine whether the titration of H-montmorillonite with sodium hydroxide, which also involves a change of dimension of the substrate due to swelling, was similar to that of humic acid.

A sample of montmorillonite from Chambers, Arizona (A.P.I. Standard No. 23, ref. 6), was converted to the H-form by treatment in 1 N hydrochloric acid by the method of Aldrich and Buchanan[7], with the omission of the final treatment with an ion-exchange resin. Care was taken to remove all traces of chloride by repeated washing and centrifugation. No aluminium was found in the supernatant water in contact with the H-montmorillonite, although this water had a pH value sufficiently low for aluminium to be in solution. This suggests that no aluminium was in the exchange positions. Titrations were made by a discontinuous method in which 65-mgm. samples of montmorillonite were suspended in 5 ml. water and increasing multiples of a 0·02-ml. increment of 0·1 N sodium hydroxide were added. The maximum volume of sodium hydroxide used was 1·2 ml. Additional water was added, when required, to make a constant volume of 6·20 ml. for each sample. The pH value was measured repeatedly until it remained constant for about six weeks. Titration curves were plotted semi-logarithmically. The volume of titrant added was entered on the logarithmic ordinate, and the pH on the linear abscissa. The result is shown in Fig. 1.

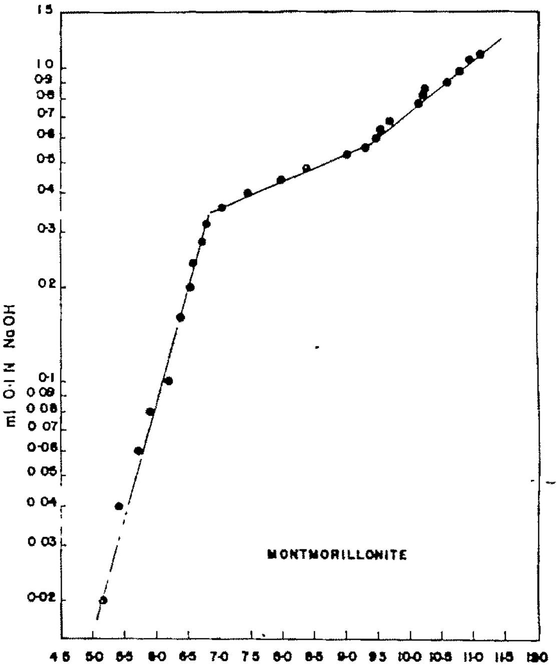

Fig. 1 Titration of H-montmorillonite with sodium hydroxide

The plot was made in this way for the reasons set out below. Gran[8] has shown that if a dibasic weak acid is titrated with a strong base the following relation holds before the first equivalence point is reached:

$$10^{-pH} = \frac{K_a}{V} (V_1 - V)$$

Here K_a is the first ionization constant of the acid, V is the volume of base added, and V_1 is the volume necessary to reach equivalence. Upon logarithmic transformation this relation yields:

$$pH = \log V - \log K_a - \log (V_1 - V)$$

This equation can be rearranged to:

$$pH = \log V - \log K_a - \log V_1 + \log (1 + \frac{V}{V_1})$$

a relation which shows that a plot of pH against $\log V$ will yield a straight line except for a deviation contributed by the last term in this equation. Evaluating $\log (1 + V/V_1)$ we find that, at the start of the titration, $V = 0$ and $\log (1 + V/V_1) = 0$, whereas at the point at which equivalence is reached, $V = V_1$ and $\log (1 + V/V_1) = \log 2 = 0{\cdot}30$. Hence, a plot of $\log V$ against pH over this region will deviate from linearity by 0·30 pH units. This small deviation may be neglected.

In the region between the first and second equivalence points, Gran[8] showed that:

$$10^{-pH} = K'_a \frac{V_2 - V}{V - V_1}$$

where K'_a is the second ionization constant of the acid and V_2 is the volume of base necessary to reach the second equivalence point. By using the same approach as before, this equation can be rewritten as:

$$pH = \log (V - V_1) - \log K'_a - \log V_2 - \log (1 + \frac{V}{V_2})$$

and it is possible to plot pH against $\log (V - V_1)$ as essentially a straight line.

A plot of $\log V$ against pH will also yield a straight line. Gran[8] has shown that, after the second equivalence point has been passed:

$$10^{pH} = \frac{C_B}{f_H K_w (V_0 + V)} (V - V_1)$$

where C_B is the concentration of base, f_H is the activity coefficient of the hydrogen ions, K_w is the ionization constant of water, and V_0 is the volume at the start of the titration. Again, it can be expected from this relation that also in this region a plot of $\log V$ against pH will yield a straight line.

If, then, $\log V$ is plotted against pH when a dibasic weak acid is titrated with a strong base, three approximately straight lines should appear. These lines should intersect at the equivalence points. A plot of V against pH, based on the discontinuous titration of H-montmorillonite showed that this is so for this clay mineral (Fig. 1). It is possible to estimate from the equivalence points in Fig. 1 that the cation exchange capacity of the first clay acid, presumably representing the interlayer (C^-) sites, is about 50 m.equiv./100 gm., and that the cation exchange capacity of the second acid, presumably representing the edge sites (E^-), is about 40 m. equiv./100 gm. The total exchange capacity of this H-montmorillonite as determined by this method is about 90 m. equiv./100 gm. The exchangeable cations remaining on this H-clay, as determined by the Association of Official Agricultural Chemists method[9], were 34 m. equiv./100 gm., and the total exchange capacity of the original montmorillonite was, therefore (90 + 34 m. equiv.)/100 gm. or 124 m. equiv./100 gm., a value comparing closely with that of ~ 120 m. equiv./100 gm. reported for this mineral[10].

A plot of $\log \alpha/(1 - \alpha)$, values for which were easily obtained after the exchange capacity was determined, against pH over the region of the neutralization of the first acid gave a straight line with a slope of 1 and an intercept of 6·4 (Fig. 2a). A few points in the region where $1 - \alpha$ approached zero could not be used in the construction of this line. It appears from the value of the intercept that the ionization constant of the first acid is $10^{-6{\cdot}4}$ and from the slope that the acid behaves as a pure weak acid upon neutralization. A similar plot over the region of the neutralization of the second acid (Fig. 2b) gave a straight line with an intercept of 8·3, indicating an ionization constant of the second clay acid of $10^{-8{\cdot}3}$ and a slope of 0·7. This shows that the clay acid deviates in its behaviour from that of pure weak acid, and that it can be described by the type of generalized Henderson–Hasselbalch equation used in the titration of polyelectrolytes. The value of n in this case, however, is smaller than 1. The reason for this deviation has not yet been explored.

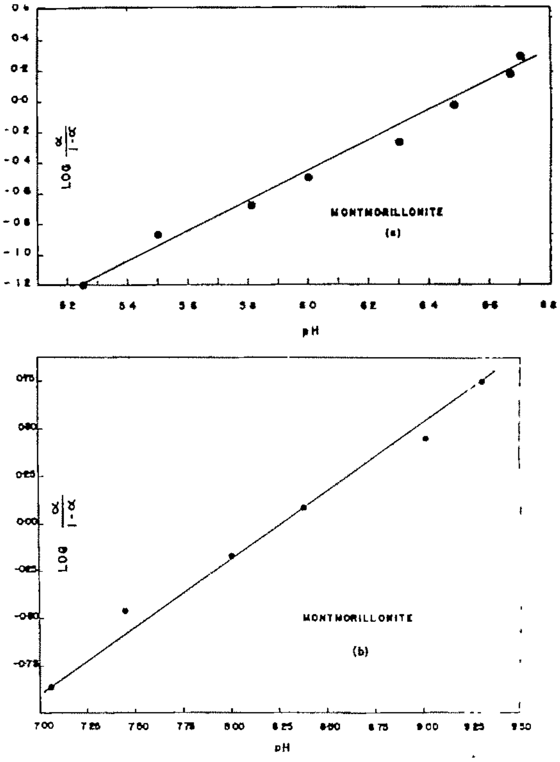

Fig. 2. (*a*) Plot of $\log \alpha/(1-\alpha)$ against pH for first clay acid; (*b*) plot of $\log \alpha/(1-\alpha)$ against pH for second clay acid

We think that plotting the results of discontinuous titrations of H-clays as described is a useful way to characterize their exchange relations. We have made similar titrations for vermiculite and 'illite'. These results, together with that for montmorillonite, will be described in a more comprehensive paper now being prepared for publication.

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[1] Marshall, C. E., "The Colloid Chemistry of the Silicate Minerals", 101 (Academic Press, New York, 1949).

[2] Garrels, R. M., and Christ, C. L., *Amer. J. Sci.*, **254**, 372 (1956).

[3] Marshall, C. E., and Bergman, W. E., *J. Phys. Chem.*, **46**, 52, 327 (1942).

[4] Blackmon, P. D., *Amer. J. Sci.*, **256**, 733 (1958).

[5] Katchalsky, A., and Spitnik, P., *J. Polymer Sci.*, **2**, 432 (1947).

[6] American Petroleum Institute Project 49, Prelim. Rep. No. 7 (1950).

[7] Aldrich, D. G., and Buchanan, J. R., *Soil Sci. Soc. Amer. Proc.*, **22**, 281 (1958).

[8] Gran, G., *Analyst*, **77**, 661 (1952).

[9] Association of Official Agricultural Chemists, "Official Methods of Analysis", 41, 7th Ed. (A.O.A.C., Washington, D.C., 1950).

[10] Higdon, W. T., and Marshall, C. E., *J. Phys. Chem.*, **62**, 1204 (1958).

CHEMICAL AND SEROLOGICAL RELATIONSHIPS OF CERTAIN BACTERIAL POLYSACCHARIDES CONTAINING SIALIC ACID

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SINCE the composition of the polysaccharide, colominic acid, obtained from an *E. coli* organism, was established[1] as poly N-acetylneuraminic acid (ovine sialic acid), reports have appeared describing other bacterial polysaccharides that contain a sialic acid[2,3]. One of these substances, the specific hapten of group *C N. meningitidis*[2], is also largely constituted of units of O-sialic acid. Thus, it appeared that colominic acid may be produced by species of bacteria other than *E. coli*. It was of interest to ascertain by agglutination tests whether the various species of bacteria known to produce polysaccharides rich in sialic acid showed any relationship to each other. In addition, a study was made of the chemical and serological properties of four polysaccharides containing sialic acid obtained from different bacteria.

All enteric bacteria were grown in sterile culture. The bottles containing 15 l. of suitable medium were incubated (with aeration) at 37° C. for 18 hr. The bacteria were killed by the addition of 100 ml. of chloroform and harvested by centrifugation. The cells were suspended in 100 ml. of water and collected as before. The washed bacteria were suspended in 100 ml. of water and lyophilized.

N. meningitidis bacteria were cultivated at 37° C. for 18 hr. on 10 sterile blood agar plates. The cells were carefully removed by flushing each plate with 5 ml. of distilled water. The cells were collected by centrifugation, washed, and lyophilized.

Sialic acid analyses were performed on the dried bacteria, employing a modification of Werner and Odin's method[4]. N-acetylneuraminic acid served as the standard and the figures recorded represent typical values, expressed as a percentage of dry weight: *Escherichia coli* 01 : *K*1 : *HNM* (0·27); *E. coli* 02 : *K*1 : *HNM* (0·46); *Neisseria meningitidis* group *C* 1908 (0·64); *N. meningitidis* group *C* 1935 (0·26); *Citrobacter freundii* (1·92); *Salmonella dahlem* (1·19); and *S. djkarta* (0·51).

Antibacterial antisera prepared against the *K* antigen of the *E. coli* strains were obtained by Edwards's procedure[5]. *N. meningitidis* antibacterial antisera were prepared by Rake's method[6]. Edwards and Ewing's method[7] was used for production of *H* antigen antibacterial antisera prepared against the *C. freundii*, *S. djkarta* and *S. dahlem* strains.

The results of the agglutination tests are given in Table 1. It is to be noted that all the bacteria show agglutination in homologous antisera. The *E. coli* 01 : *K*1 : *HNM* and the group *C N. meningitidis* 1908 strains are unrelated by cross agglutination tests to each other or to any of the other strains examined. The cross agglutination tests reveal a relationship between the two *Salmonella* strains and

Table 1. Agglutination of Bacteria containing Sialic Acid in Homologous and Heterologous Antisera

Organism tested	Antibacterial antiserum	Dilution of antiserum										
		1/5	1/10	1/20	1/40	1/80	1/160	1/320	1/640	1/1,280	1/2,560	1/5,120
S. djkarta	*S. djkarta*	4+	4+	4+	4+	4+	4+	4+	4+	4+	1+	±
S. dahlem		4+	4+	4+	4+	4+	4+	2+	1+	±		
C. freundii		4+	4+	4+	4+	3+	2+	0	0	0	0	0
E. coli 01 : *K*1 . *HNM*		0	0	0	0	0	0	0	0	0	0	0
N. meningitidis group *C* 1908		0	0	0	0	0	0	0	0	0	0	0
S. djkarta	*S. dahlem*	4+	4+	4+	4+	4+	4+	4+	3+	2+	0	0
S. dahlem		4+	4+	4+	4+	4+	4+	4+	4+	4+	3+	2+
C. freundii		4+	4+	4+	4+	4+	2+	0	0	0	0	0
E. coli 01 : *K*1 : *HNM*		0	0	0	0	0	0	0	0	0	0	0
N. meningitidis group *C* 1908		0	0	0	0	0	0	0	0	0	0	0
S. djkarta	*C. freundii*	4+	4+	4+	4+	4+	4+	4+	4+	4+	1+	0
S. dahlem		4+	4+	4+	4+	4+	4+	4+	4+	4+	1+	0
C. freundii		4+	4+	4+	4+	4+	4+	4+	4+	4+	4+	4+
E. coli 01 : *K*1 : *HNM*		0	0	0	0	0	0	0	0	0	0	0
N. meningitidis group *C* 1908		0	0	0	0	0	0	0	0	0	0	0
S. djkarta	*E. coli* 01 : *K*1 : *HNM*	0	0	0	0	0	0	0	0	0	0	0
S. dahlem		0	0	0	0	0	0	0	0	0	0	0
C. freundii		0	0	0	0	0	0	0	0	0	0	0
E. coli 01 : *K*1 : *HNM*		4+	4+	4+	4+	3+	2+	+	0	0	0	0
N. meningitidis group *C* 1908		0	0	0	0	0	0	0	0	0	0	0
S. djkarta	*N. meningitidis* group *C* 1908	0	0	0	0	0	0	0	0	0	0	0
S. dahlem		0	0	0	0	0	0	0	0	0	0	0
C. freundii		0	0	0	0	0	0	0	0	0	0	0
E. coli 01 : *K*1 : *HNM*		0	0	0	0	0	0	0	0	0	0	0
N. meningitidis group *C* 1908		4+	4+	4+	4+	4+	4+	2+	2+	1+	0	0

Table 2. Composition of Polysaccharides containing Sialic Acid obtained from Various Bacteria

Source organism	Substance isolated	Empirical formula	Elemental analysis					Per cent sialic acid
			C	H	N	O	CH_3CO	
E. coli 01 : *K*1 : *HNM*	Colominic acid	$(C_{11}H_{19}NO_9)_n$	42 83	6·46	4·79	45·97	14·45	99 0
E. coli 02 : *K*1 : *HNM*	Colominic acid	$(C_{11}H_{19}NO_9)_n$	43·75	6 36	4·71	45·18	13 88	98·5
N. meningitidis group *C* 1935	Group *C* hapten	$(C_9H_{16}NO_8)_n$	41·80	6 66	5·32	46·24	11·62	85 0
C. freundii	Polysaccharide	$(C_{15}H_{28}N_2O_{11})_n$	44·19	7·16	6·86	41 79	10 57	36·2

between each of these with the *C. freundii* organism. Cross agglutination tests, not recorded here, show that *E. coli* 02 : *K*1 : *HNM* is related to *E. coli* 01 : *K*1 : *HNM* and to no other strain listed in Table 1. Furthermore, a group *C N. meningitidis* strain known as 1935 is related only to the group *C N. meningitidis* 1908 strain and to none other listed here. Therefore, on the basis of these tests the above bacteria which produce polysaccharides that contain sialic acid can be divided into three groups.

Polysaccharides containing sialic acid were isolated from members of each group. Colominic acid was isolated from the culture media of *E. coli* 01 : *K*1 : *HNM* and 02 : *K*1 : *HNM* bacteria by a previously reported procedure[8]. This procedure, with a few modifications, was employed for the preparation of a new polysaccharide that contains sialic acid isolated from the culture medium of the *C. freundii* organism.

The results of the chemical analyses are recorded in Table 2. It is to be noted that colominic acid isolated from either *E. coli* 02 : *K*1 : *HNM* or 01 : *K*1 : *HNM* bacteria has similar elemental compositions and sialic acid content with one acetyl group for each nitrogen atom. Furthermore, identical infra-red absorption spectra and optical rotation values are obtained. N-acetylneuraminic acid is the only substance found in hydrolysates of either polymer. The calculated empirical formulæ support the belief that O-sialic acid ($C_{11}H_{19}NO_9$) is the sole chemical constituent present in these polysaccharides.

Group *C* hapten isolated from group *C N. meningitidis* 1935 strain contains about 85 per cent sialic acid. An empirical formula $(C_9H_{16}NO_8)_n$ also fits the elemental analytical data reported[2].

The group *C* hapten 1908 used in the present study contains 70·4 per cent sialic acid and a small amount of protein[9]. Hexosamines[10] are absent. The results of these tests indicate that a portion of the group *C* hapten is undetermined.

The *C. freundii* polysaccharide contains 36 per cent sialic acid and one of the nitrogen atoms is believed to be acetylated. Hexuronic acids are absent and only trace amounts of protein can be detected. About 21 per cent of hexosamine is present. Phosphorus is absent. Thus, half this polysaccharide, rich in nitrogen, remains to be characterized.

All four polysaccharides readily decompose upon heating in dilute mineral acid to form humin. Colominic acid does not form precipitins with antibacterial antisera prepared against *E. coli* 01 : *K*1 : *HNM*, 02 : *K*1 : *HNM* or group *C N. meningitidis* 1908 and 1935 bacteria. The group *C* hapten yields a precipitin with the group *C N. meningitidis* antibacterial antisera in a dilution of 1 part in 10^6 but not with the *E. coli* 01 : *K*1 : *HNM* or 02 : *K*1 : *HNM* antisera. Incubation of colominic acid with *N. meningitidis* 1908 or 1935 antisera fails to block precipitin formation by the group *C* hapten. Incubation of group *C* hapten with *E. coli* 01 : *K*1 : *HNM* or 02 : *K*1 : *HNM* antibacterial antiserum followed by the addition of *E. coli* 01 : *K*1 : *HNM* or 02 : *K*1 : *HNM* bacteria does not inhibit agglutination of the cells.

Hæmagglutination tests employing group *C* hapten adsorbed to human O erythrocytes in group *C N. meningitidis* 1908 or 1935 antisera show strong hæmagglutination of the cells. However, no hæmagglutination occurs with *E. coli* 01 : *K*1 : *HNM* or 02 : *K*1 : *HNM* antibacterial antisera. Incubation of colominic acid with either *N. meningitidis* antiserum prior to the addition of the coated erythrocytes fails to block hæmagglutination of the cells. Therefore, it must be concluded that on the basis of the serological tests the group *C* hapten obtained from group *C N. meningitidis* organisms is unrelated to colominic acid obtained from *E. coli* bacteria despite the chemical similarity of the two materials. The absence of a serological relationship suggests differences in chemical linkage between the constituent units.

The *C. freundii* polysaccharide gives a precipitin with antibacterial antisera prepared against *C. freundii*, *S. djkarta* and *S. dahlem* bacteria.

Up to the present time the sialic acids have been found only in Gram-negative organisms. Three polysaccharides containing sialic acid obtained from such organisms—colominic acid from *E. coli*, group *C* hapten from *N. meningitidis* and a previously undescribed material from a *C. freundii* organism—were found to contain different amounts of sialic acid and to be unrelated on the basis of serological tests. Colominic acid remains as a unique material produced only by *E. coli* organisms which possess a *K*1 antigenic serotype[11]. Precipitin tests indicate that a polysaccharide rich in sialic acid obtained from a *C. freundii* organism is related to a similar material produced by *S. dahlem* and *S. djkarta* bacteria. Thus, the occurrence of the sialic acids in various bacterial polysaccharides in different chemical combinations indicates that these monosaccharides are common structural constituents of cells.

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[1] Barry, Guy T., *Science*, **126**, 1230 (1957).

[2] Watson, Glenn R., Marinetti, Guido, V., and Scherp, Henry W., *J. Immunol.*, **81**, 331 (1958).

[3] DeWitt, C. W., and Rowe, J. A., *Nature*, **184**, 381 (1959).

[4] Werner, I., and Odin, L., *Acta Soc. Med Upsaliensis*, **57**, 230 (1952).

[5] Edwards, P. R., *Pub. Health Rep.*, **66**, 837 (1951).

[6] Rake, G., *J. Exp. Med.*, **57**, 561 (1933).

[7] Edwards, P. R., and Ewing, W. H , "Ident. of Enterobact." (Burgess Pub. Co., Minneapolis, Minn., 1955).

[8] Barry, Guy T., *J. Exp. Med.*, **107**, 507 (1958).

[9] Kunkel, H. G., and Tiselius, A., *J. Gen. Physiol.*, **35**, 89 (1951).

[10] Sorensen, M., *C.R. Trav. Lab. Carlsberg*, **22**, 487 (1938).

[11] Barry, Guy T., *Nature*, **183**, 117 (1959).

LETTERS TO THE EDITORS

RADIOPHYSICS

Constant Ionosphere Height for Audio-frequency Propagation

THE wave-form of an atmospheric, resulting from a distant lightning flash to Earth, has variously been interpreted in terms of pulse reflexion between parallel or concentric conducting Earth and ionospheric layers[1,2] or as pulse dispersion during propagation in the region between these same layers[3]. Recent papers[4,5] have discussed the equivalence of these approaches. In each case the Earth–ionosphere separation and the storm distance are the principal determinants of the positions of the peaks of the trace. Within certain limits these propagation parameters may be estimated from the observed wave-forms.

Smooth type wave-forms[5] of distant Atlantic origin, received both by day and night, have recently been studied extensively at Nottingham. Wave-form analyses of 70 traces, using wave-guide concepts, together with distance estimates in the range 1,750–5,000 km., kindly provided by the Meteorological Office Sferics network, have permitted estimation of the effective ionosphere height both by day and for several hours after sunset in the case of winter-time storms. A constant ionosphere height of 83 ± 2 km. irrespective of the time of day was the most reasonable conclusion from the results available, with no significant variations from this mean value for individual recording periods and no suggestion of a systematic variation with the onset of night-time conditions even up to 4 hr. after local sunset. This suggests a fundamental difference between smooth and reflexion type wave-forms, for the latter of which various workers[1,2] have reported an increase in effective height at night.

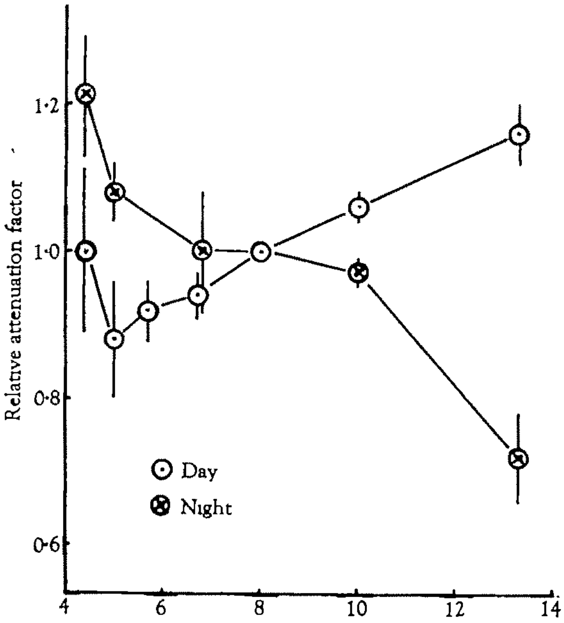

Fig. 1. Attenuation factors. (Fractional changes per 1,000 km. relative to 8 kc./s.)

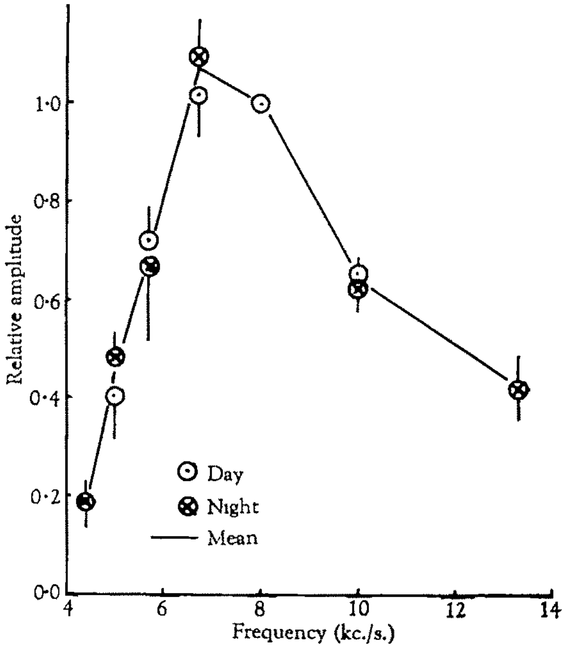

Fig. 2. Average source spectra

An earlier paper[5] has discussed the derivation of relative attenuation coefficients for propagation and the mean amplitude-spectrum at the source from statistical analysis of the oscillation amplitudes throughout the observed wave-forms. For a group of 150 apparently regular smooth type wave-forms, it was found that attenuation factor per 1,000 km. relative to that at 8 kc./s. varied little throughout the frequency-range 4–14 kc./s. A slight systematic increase with frequency was noted by day and a less precise and regular decrease at night (Fig. 1). The corresponding source spectra were indistinguishable (Fig. 2).

The very slight diurnal variation of attenuation in the appropriate frequency band, while unexpected when comparison is made with previous atmospherics measurements[6], might have been predicted from the very close similarity of day- and night-time forms of this restricted group of smooth type wave-forms. The identity of the day and night source spectra provides gratifying confirmation of the analysis and the significance of its conclusions.

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Jan. 20.

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[1] Schonland, B. F. J., Elder, J. S., Hodges, D. B., Phillips, W. E., and van Wyk, J. W., *Proc. Roy. Soc.*, A, **176**, 180 (1940).

[2] Caton, P. G. F., and Pierce, E. T., *Phil. Mag.*, **43**, 393 (1952).

[3] Hales, A. L., *Proc. Roy. Soc.*, A, **193**, 60 (1948).

[4] Budden, K. G., *Phil. Mag.*, **42**, 1 (1951).

[5] Hepburn, F., *J. Atmos. Terr. Phys.*, **14**, 262 (1959).

[6] Bowe, P. W. A., *Phil. Mag.*, **42**, 121 (1951).

Measurement of Solar and Diurnal Effects in the High Atmosphere by Artificial Satellites

IN a previous investigation[1] atmospheric densities had been derived from the orbital elements[2] of the satellites 1958 β 2, 1958 α, 1958 δ and others, and reduced to the mean altitude of the perigee by means of the Kallmann–Juncosa[3] model. A good correlation exists between atmospheric fluctuations and the 20-cm. solar flux curve[4]. This is apparent not only from the 'monthly' fluctuations[5] but also from long-period variations extending over several months. For altitudes of 660, 350 and 210 km. the relation between density and solar flux is approximately linear for values of solar flux S between 100 and 240×10^{-22} W./m.^{2} c./s. To demonstrate the effect it was necessary to separate quantitatively the suspected diurnal fluctuations and the fluctuations caused by variable solar radiation. A preliminary investigation was carried out using the geocentric angular distance perigee–Sun which had already been employed by Jacchia (see ref. 2) for comparison with the satellite data. The angular distance proved very useful so long as no separation of the diurnal and terrestrial effects was possible. One expected, however, the atmospheric density to be dependent on $\Delta\alpha = \alpha_\pi - \alpha_\odot$, $\Delta\delta = \delta_\pi - \delta_\odot$ and the geocentric latitude ψ of the perigee (where α is right ascension, δ is declination, π is perigee, $\odot$ is Sun).

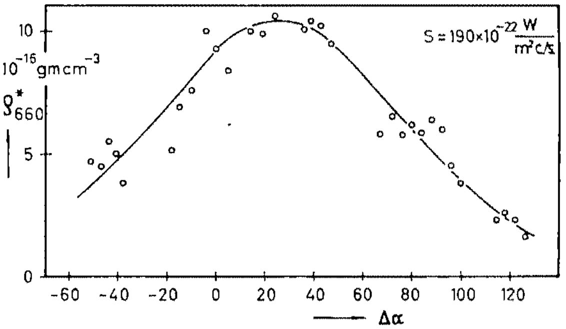

Fig. 2. Dependence of atmospheric density on $\Delta\alpha = \alpha_\pi - \alpha_\odot$ in the equatorial zone (diurnal effect)

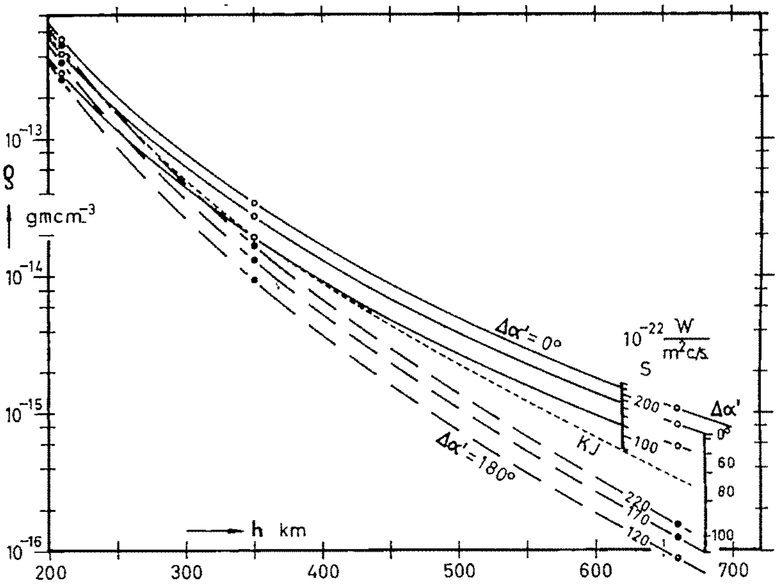

Fig. 3. Model of air density derived from the orbital elements of the satellites 1958 β 2, α and δ 2 with solar and diurnal effects ($\Delta\alpha' = \Delta\alpha - 25° = 0°$ and 180° corresponding to 13h. 40m. and 1h. 40m. local time). The short dotted line (*KJ*) represents the model by Kallmann and Juncosa (ref. 3)

A separation of the diurnal effect (dependence on $\Delta\alpha$) is now possible for the satellite 1958 β 2, for which the best observational data are available. The result is presented in Figs. 1 and 2. For this purpose the atmospheric density data were first reduced to a standard value of the solar 20-cm. radiation ($S = 190 \times 10^{-22}$ W./m.^{2} c./s.). This was possible because of the linear relation between atmospheric density and 20-cm. radiation which had already been found[1]. The resulting values were drawn in a $\Delta\alpha$, $\Delta\delta$ co-ordinate system and the lines of constant density determined. They are shown in Fig. 1 for values of 2, 4, 6, 8 and 10×10^{-16} gm. cm.$^{-3}$. For simplification in the diagram, the individual values are represented in groups (see right-hand corner of diagram). The dotted line represents $\delta_\pi = 0$ ('perigee above the equator'). The lines of constant density are strongly influenced by the scattering of the individual values. For this reason only the general appearance of the curves can be considered reliable. From this appearance one can deduce two effects: (1) a seasonal effect (dependent on the declination of the Sun) with a maximum value of atmospheric density at $\Delta\delta = 0$; and (2) a terrestrial effect with a maximum value of the density above the equator. The two effects are superposed.

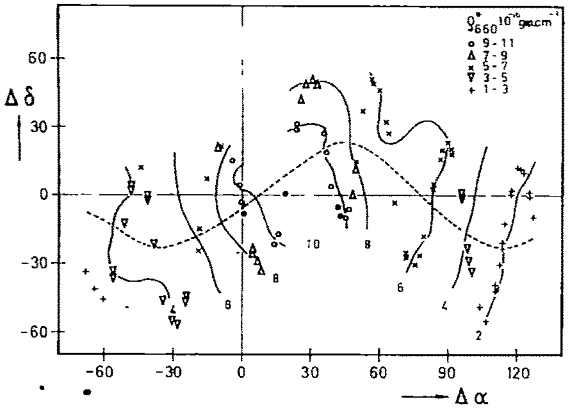

Fig. 1. Atmospheric density at an altitude of 660 km. in a co-ordinate system centred at the subsolar point. $\Delta\alpha = \alpha_\pi - \alpha_\odot$, $\Delta\delta = \delta_\pi - \delta_\odot$ (α, right ascension; δ, declination; π, perigee; $\odot$, Sun). The values of atmospheric density are reduced to a flux of solar 20-cm. radiation of $S = 190 \times 10^{-22}$ W./m.^{2} c./s.

Most conspicuous is the strong diurnal variation of $\Delta\alpha$ (Fig. 2) and the phase shift of about 25° corresponding to a maximum value of atmospheric density around 13h. 40m. true local time. The value of the delay is somewhat uncertain[1]. Using a simplified method for analysing the results, we found[1] a shift of 16°. Wyatt[6] found 23° (1h. 30m.) and an asymmetric diurnal variation. The asymmetry may result from the fact that in his derivation all values were included independent of variations in $\Delta\delta$ and ψ. If only values of the density from a narrow zone around the equator are used with both

$|\psi| < 30°$ and $|\Delta\delta| < 30°$, a much smaller scattering of the results is found. In our earlier paper (ref. 1, Fig. 3), some points did not fit the general curve. It was found that all of them represent density values for which $|\Delta\delta| > 30°$.

Using atmospheric densities as derived in our earlier communication[1] for altitudes of 210, 350 and 660 km. we obtain a density model with solar and diurnal fluctuations as shown in Fig. 3. The scale-heights of the atmosphere according to Kallmann and Juncosa[3] on which the computations were originally based are eliminated from the final result by means of an iteration process (see ref. 1).

In Fig. 3 the three upper curves (solid lines) are valid for $\Delta\alpha' = \Delta\alpha - 25° = 0°$ (13h. 40m. local time); the three lower (dotted) curves are valid for $\Delta\alpha' = 180°$ (extrapolated values). In each group the lowest of the three curves corresponds to a value of $S = 120$, the middle curve to $S = 170$ and the upper curve to $S = 220 \times 10^{-22}$ W./m.^{2} c./s. of the solar 20-cm. radiation. The large density fluctuation between day and night suggests a daily up and down movement of the atmosphere[6].

A layer of constant atmospheric density, for example, would rise from 520 to 660 km. within 6h. according to Fig. 2. The resulting velocities in the vertical direction are about 25 km./hr. The air movements decrease with decreasing altitude and at 200 km. become insignificant compared with the fluctuation caused by solar radiation. The latter must originate in a layer below 200 km. This suggests that the absorption of solar X-ray radiation in the E-layer of the ionosphere is responsible for them. The scale height $H = \frac{RT}{gM}$ (where R is the gas constant, T is temperature, g is acceleration of gravity, M is mean molecular weight) of the atmosphere and thus the ratio T/M changes strongly with the time of the day but very little with the flux of 20-cm. radiation.

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[1] Priester, W., and Martin, H. A., *Mitt. Sternwarte Bonn*, 29 *Forschungsber.* NRW (Westdeutscher Verlag, Köln-Opladen) (in the press).

[2] Jacchia, L. G., Smith. Astrophys. Obs. Spec. Rep. 29 (1959). Space Track Control Center, Bull. Orbital Elements 1958, 1959, Smith. Astrophys. Obs., Bull. Orbital Elements 1958, 1959, Paetzold, H. K., *Planet. Space Sci.*, **1**, 115 (1959).

[3] Kallmann, H. K., and Juncosa, M. L., Rand Report Oct. 30 (1958).

[4] Heinrich-Hertz-Institut, Berlin-Adlershof, Beobachtungen 1957-59.

[5] Jacchia, L. G., Harvard Obs. Announcement Card 1423 (1959). Priester, W., *Mitt. Sternwarte Bonn* 24; *Naturwiss.*, **46**, 197 (1959).

[6] Wyatt, S. P., *Nature*, **184**, 351 (1959).

PHYSICS

Natural Radioactivity of Samarium and Neodymium

There seems to exist a discrepancy between the measured value of the alpha-decay half-life of natural samarium (samarium-147) and the theoretical half-life calculated from its measured alpha energy[1]. This may be due to the fact that in most ion-chamber measurements of samarium-147, heavy natural alpha-emitters with much higher energies have been used as standards, which leads to great difficulties because of the non-linearity of the ionization process.

In this investigation thin samples containing about 50 μgm./cm.^{2} of natural samarium, 0·3 μgm./cm.^{2} of natural uranium and 0·5 μgm./cm.^{2} of elemental boron were analysed in a grid-type ionization chamber in the presence of a thermal neutron flux of $1·5 \times 10^3$ n/cm.^{2} obtained from a polonium–beryllium source.

The neutrons absorbed by boron gave rise to alpha-particles with energies of 1·797 and 1·474 MeV.[2], respectively, which were recorded photographically together with the samarium and uranium alpha-particles for the purposes of calibration[3].

By the use of exposures several days long, spectra containing 10^4–10^5 pulses were obtained and analysed with a spectrophotometer. The result for the alpha energy of samarium-147 is:

$$E = 2·20 \pm 0·03 \text{ MeV.}$$

which is somewhat higher than the value quoted by W. Jesse and J. Sadauskis[4].

The specific activity of samarium-147 was measured by counting the alpha-particles from 28·9 mgm. of samarium which were vacuum-evaporated on a source plate of diameter 200 mm. Taking into account self-absorption and back-scattering, a value of 7·00 ± 0·3 counts/min./mgm. of natural samarium is obtained, which corresponds to a half-life of $(1·14 \pm 0·05) \times 10^{11}$ yr. for the isotope 147.

An attempt to detect the natural alpha activity of neodymium was made using a 200-mm. source-plate on which 36 mgm. of neodymium were evaporated. In spite of appreciable thorium contamination a weak line was observed at (2·0 ± 0·1) MeV., in agreement with investigations made by the emulsion method[5].

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[1] Segré, E., "Experimental Nuclear Physics", **3**, 140 (John Wiley and Sons, 1959).

[2] Chao, C. Y., Lauritsen, C. C., and Tollestrup, A. V., *Phys. Rev.*, **76**, 586 (1949).

[3] For a detailed report on the apparatus, see Karras, M., "On the Natural Radioactivity of Samarium" (to be published).

[4] Jesse, W. P., and Sadauskis, J., *Phys. Rev.*, **78**, 1 (1950).

[5] Waldron, E. C., Schultz, V. A., and Kohman, T. P., *Phys. Rev.*, **93**, 254 (1954).

Evaporation Figures on the Surface of Sodium Chloride Crystals

Figures which appear during evaporation on the surface of sodium chloride have already been reported by E. Kern and H. Pick[1], S. Amelinckx and E. Votava[2], and others. Here we report on figures obtained during evaporation experiments which recall similar figures obtained on the surface, for example, of silicon carbide by A. R. Verma[3] and on sodium chloride by Z. Morlin[4].

The annealing of the sodium chloride plates (15 mm. × 5 mm. × 2 mm.) was carried out near the melting point for 4–5 days in a crucible made from sodium chloride single-crystal and covered with a single-crystal plate. Besides the simple-concentric circles seen in Fig. 1 (the small spots on the figure are etch-pits revealed by glacial acid), figures of sharp characteristic contours were obtained (Fig. 2). These may represent Frank–Read sources, which appear at the edges of the sodium chloride plates,

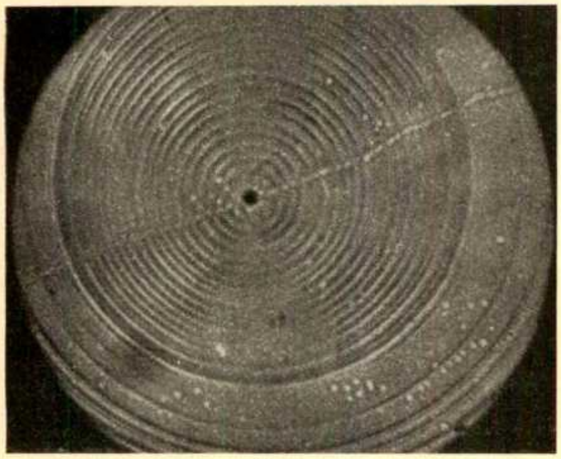

Fig. 1

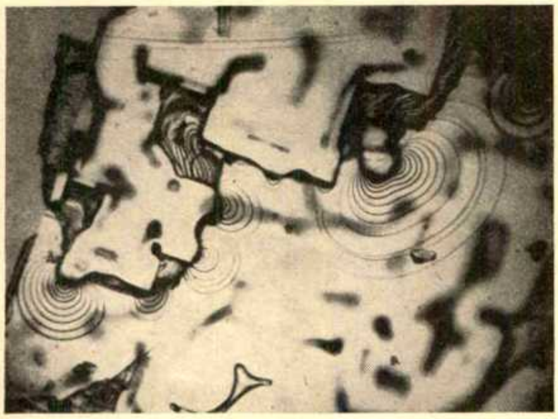

Fig. 2

especially on the edge of the surface where the two sodium chloride plates lay on each other during the evaporation.

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[1] Kern, E., and Pick, H., *Z. Physik*, **134**, 610 (1953).

[2] Amelinckx, S., and Votava, E., *Naturwiss.*, **18**, 422 (1954).

[3] Verma, A. R., "Crystal Growth and Dislocations" (Butterworths Publications, London, 1953).

[4] Morlin, Z., *Nature*, **183**, 1319 (1959).

Exo-electron Emission and the Optical Properties of Oxide-coated Metal Surfaces

EVIDENCE suggests that exo-electron emission from ionic crystals is associated with colour centres and kindred imperfections which result in energy-levels in the forbidden band of the crystal[1]. Grunberg and Wright[2] found very strong exo-electron emission from abraded aluminium, zinc and magnesium irradiated with light at 4700 Å. The tendency of these elements to form metal excess oxides led Grunberg and Wright to suggest that these may be present in the oxide oxygen ion vacancies occupied by two electrons called, by analogy with the alkali halides, F'-centres. The excitation of the F'-centres by light of 4700 Å. was suggested as an important process in the mechanism of exo-electron emission from metals.

The method of preparation of metal surfaces is known to affect the experimentally determined optical constants, no doubt due to differences in structure of the surface layers. The oxide film on a mechanically polished metal surface would probably contain large numbers of the proposed F'-centres, which might be expected to increase the dielectric constant of the film in the region of 4700 Å. with a consequent alteration in the measured values of the optical constants.

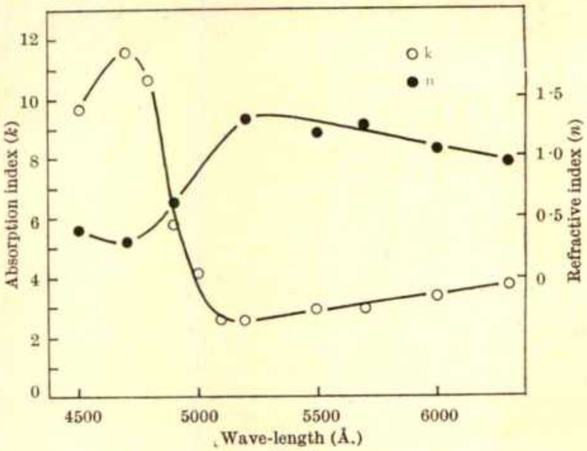

Fig. 1

The optical constants of mechanically polished aluminium and zinc have been measured for various wave-lengths in the visible region by the Drude method. Fig. 1 illustrates the variation in the absorption index, k, and the refractive index, n, for aluminium, showing a considerable increase and decrease, respectively, in the vicinity of 4700 Å. A similar result was obtained with zinc. Further work, at present in progress, is, however, required to establish a definite relationship with the proposed F'-centres.

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[1] Bohun, A., *Czech. J. Phys.*, **3**, 2 (1953).

[2] Grunberg, L., and Wright, K. H. R., *Proc. Roy. Soc.*, A, **232**, 423 (1955).

Ratio of Nucleon Mass and Electron Mass

WITH reference to the experimental value 1840 for the ratio of nucleon and electron mass, mentioned by Prof. A. J. Rutgers in *Nature* of September 19, p. 1959, it may be noted that the ratio of proton mass and electron mass is given by the exponential equation:

$$\frac{m_p}{m_e} = \frac{1}{2}\exp(-1) \times 10^4 = 1839 \cdot 39$$

More generally, the rest mass of an elementary particle, relative to the rest mass of the electron, is $m/m_e = k\exp(-x)$, and since $\exp(-x)$ is a 'die-away factor', this formula represents the tendency of the mass of an elementary particle to decrease. For all elementary particles except the electron with minimum rest mass, and the proton with relative rest mass $m_p/m_e = k\exp(-1)$, the tendency to decrease of rest mass has been demonstrated experimentally.

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Observation of Dislocations in Non-metallic Layer Structures

TRANSMISSION electron microscopy of thin foils, as developed by Hirsch, Whelan and co-workers[1], has become an important tool in studying the static[1,2] and kinetic[3] behaviour of dislocations in metals. Very few observations on non-metals have been published as yet. We have applied this technique to a number of non-metallic layer structures. One has then the advantage that very thin cleavage flakes can easily be prepared and, moreover, that the main glide plane usually coincides with the cleavage plane. In such cases extensive dislocation patterns will appear in the plane of observation. In particular, we have studied in some detail the following substances : bismuth telluride and antimony telluride, graphite, muscovite and talc. A few examples of patterns observed are reproduced in Figs. 1–3.

The c-plane of Bi_2Te_3 and Sb_2Te_3 is a glide plane and cleavage plane at the same time. Extensive hexagonal grids can be observed in both substances ; Fig. 1 gives an example observed in Bi_2Te_3 ; very similar patterns are observed in Sb_2Te_3. For both substances a dissociation into partials is to be expected since glide occurs between close-packed planes. However, in both substances the basal dislocations generally appear undissociated with the resolving power available with the standard Philips microscope. In Bi_2Te_3, there is an indication that they may be dissociated over a small distance. Extensive movement of dislocations in the c-plane, and in pyramidal glide planes, giving rise either to elimination of dislocations or to the formation of networks, has been observed. The driving force for this process is heating of the foil by the electron beam, and the resulting temperature gradients. Hexagonal networks were also found in the basal plane of muscovite. It was found that heating with the electron beam gave rise to the 'precipitation' of gases into small bubbles located on dislocations (Fig. 2).

In graphite and talc the dislocations are found to be split into partials. The phenomenon is especially striking in talc where the equilibrium separation turns out to be approximately equal to $0 \cdot 2\mu$, as can be judged from Fig. 3. This implies a very small stacking fault energy. This very low value of the stacking fault energy should be representative for a class of clay minerals ; it may explain why stacking faults occur so frequently in these substances. Fig. 3 shows dislocation ribbons and networks containing contracted and extended nodes somewhat similar to those observed in face-centred metals[2].

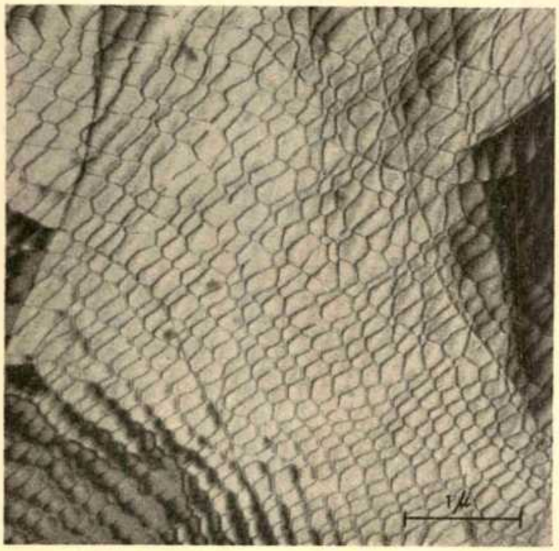

Fig. 1. Dislocation network in Bi_2Te_3. Note that certain segments of dislocations and certain node-points have left the foil

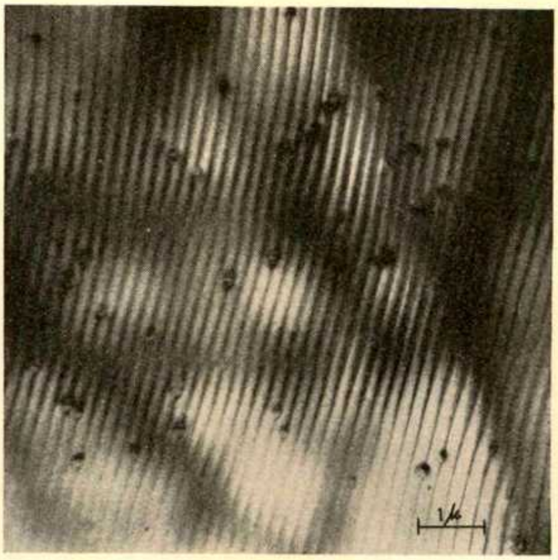

Fig. 2. Gas bubbles formed on dislocations in muscovite after irradiation with the electron beam

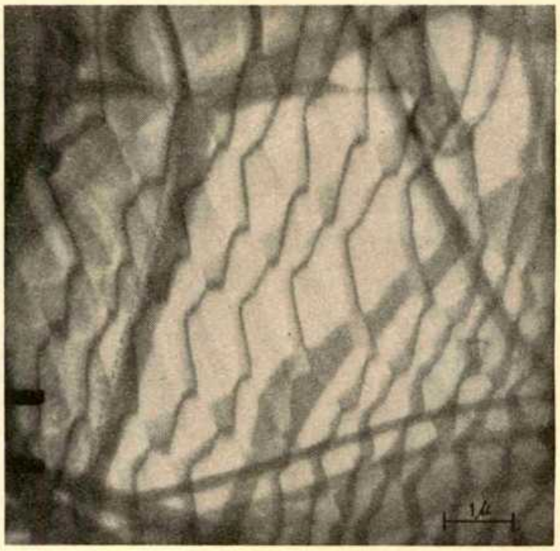

Fig. 3. Widely extended dislocations in the basal plane of talc. Note the formation of extended and contracted nodes

The dislocations in talc and in related clay minerals are of particular interest since, as a consequence of the hexagonal ring structure of the planes between which glide takes place, dislocations, split into four or six partials, are to be considered, giving rise to typical features not found in the face-centred metals.

A more detailed account of these observations will be published elsewhere.

We are indebted to Mr. J. Goens, director of the Centre d'Etude de l'Energie Nucléaire, Mol, for permission to publish this communication. We also wish to thank Mr. Nicasy for his skilful preparation of specimens and Mr. Beyens for the photographic work.

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[1] For a review, see the contributions by Hirsch, P. B., and Whelan, M. J., *J. Inst. Metals*, **4**, 385 (1959).

[2] Whelan, M. J., *Proc. Roy. Soc.*, A, **249**, 114 (1958).

[3] Berghezan, A., and Fourdeux, A., *C.R. Acad. Sci.*, **248**, 1333 (1959).

CHEMISTRY

Two New Complex Calcium Ferrite Phases

THE system CaO–FeO–Fe_2O_3 has been the subject of many investigations during the past decade, especially by Italian research workers studying the reduction of calcium ferrites and other ferrite phases of interest in cement chemistry. The phases CaO.FeO.Fe_2O_3 and CaO.3FeO.Fe_2O_3 have been identified by Burdese[1]. No ternary phases seem to have been observed in the sub-system hematite–dicalcium ferrite–magnetite. The purpose of this communication is to report the finding of two new ternary phases in this part of the system.

In a preliminary study of the systems Fe_2O_3–$CaFe_2O_4$–Fe_3O_4 and $CaFe_2O_4$–$Ca_2Fe_2O_5$–Fe_3O_4 several compositions were prepared within each triangle. Varying proportions of synthetic dicalcium ferrite, hematite and magnetite powders were mixed and small pellets were pressed. These were sealed in evacuated silica capsules, which were heated at 1,000° C. for 1–10 days and then quenched in water. The pellets were examined magnetically, microscopically, by X-rays, and analysed chemically. It soon became evident that two new ternary phases exist, located one in each of the composition triangles mentioned. The two phases appear to have the following approximate compositions: 3CaO.FeO.7Fe_2O_3 located in the Fe_2O_3–$CaFe_2O_4$–Fe_3O_4 system and 4CaO.FeO.4Fe_2O_3 in the $CaFe_2O_4$–$Ca_2Fe_2O_5$–Fe_3O_4 system. Samples having compositions close to these formulæ were non-magnetic and appeared mainly as single phases under a metallographic microscope. The stoichiometric compositions of the two new ternary phases are 24·0 per cent CaO, 7·7 per cent FeO and 68·3 per cent Fe_2O_3 for 4CaO.FeO.4Fe_2O_3 and 12·4 per cent CaO, 5·3 per cent FeO and 82·3 per cent Fe_2O_3 for 3CaO.FeO.7Fe_2O_3. Table 1 lists the *d*-spacings in Å. units and the relative intensities for the two phases as determined by a recording X-ray diffractometer.

The pattern of 3CaO.FeO.7Fe_2O_3 is strikingly similar to that reported[2,3] for calcium diferrite (CaO.2Fe_2O_3), suggesting that this ternary phase may be a solid solution between magnetite and calcium diferrite in analogy with other limited solid solutions in the system CaO–FeO–Fe_2O_3. Phase relationships and other data for the system hematite–magnetite–dicalcium ferrite will be reported when the current study of the oxygen potential for this system has been completed.

Table 1

4CaO.FeO.4Fe_2O_3 *d*	*I*/*I*$_0$	3CaO.FeO.7Fe_2O_3 *d*	*I*/*I*$_0$
11·3	25	15·7	20
5·6	35	5·2	10
3·74	10	4·9	25
2·99	30	3·11	20
2·81	100	2·97	40
2·67	15	2·92	15
2·52	15	2·67	40
2·49	15	2·62	35
2·29	20	2·58	100
2·24	15	2·54	30
2·06	20	2·15	20
1·84	15	1·99	15
1·73	15	1·84	15
1·58	10	1·70	20
1·50	10	1·56	20
		1·48	20

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[1] Burdese, A., *Met. Ital.*, **44**, 343 (1952).

[2] Edstrom, J. O., *Jernkont. Ann.*, **140**, 101 (1956).

[3] Phillips, B., and Muan, A., *J. Amer. Ceram. Soc.*, **41**, 445 (1958).

Effect of High-Energy Radiation on Mixtures of Vinyl or Related Polymers with Unsaturated Compounds

A MECHANISM has been recently proposed[1] to account for the behaviour of polypropylene on irradiation, in which the formation of a cross-link between molecules involves reaction between an activated centre in the polymer and a chain fragment containing a vinylidene double bond. This suggests that the cross-linking induced by radiation would be enhanced by the incorporation of unsaturated additives into this polymer. Considerable enhancement of cross-linking has been claimed to occur in the irradiation of 'solid solutions' of polyvinyl chloride in di- and tri-allyl esters[2].

Consideration of the above prompted an investigation into the effect of unsaturated compounds on addition polymers in general and on polymers of the vinyl family in particular. Compounds chosen to represent three classes of unsaturation were diethyl maleate, dibutyl itaconate and triallyl cyanurate. To ensure that any enhancement would be detected, it was desirable to incorporate into the polymers as much additive as possible. However, for ease of interpretation, the concentration should lie within the range in which any reaction with the polymers could be regarded as the result of an indirect effect of the radiation on the additive. An additive concentration of 10 per cent was chosen as a reasonable compromise.

The preliminary results of this survey are presented in Table 1, the criterion for enhancement of cross-linking being the variation of sol fraction with dose for the polymer plus additive when compared with that of the polymer alone (in this table 'No' indicates

Table 1

Polymer	Enhancement of cross-linking in the presence of Diethyl maleate	Dibutyl itaconate	Triallyl cyanurate
Polyethylene	None	None	Slight
Polypropylene	Slight	Slight	Good
Poly*iso*butylene	No	No	No
Polyvinyl alcohol	No	No	Good
Polyvinyl formal	Good	No	Good
Polyvinyl acetate	Slight	Slight	Good
Polyvinyl methyl ether	No	No	No
Polyvinyl chloride	Slight	No	Good
Polyvinyl pyrrolidone	No	No	Good
Polystyrene	No	No	Good
Polyvinyl carbazole	No	No	Good
Polymethyl acrylate	None	None	Slight
Polymethyl methacrylate	No	No	Good

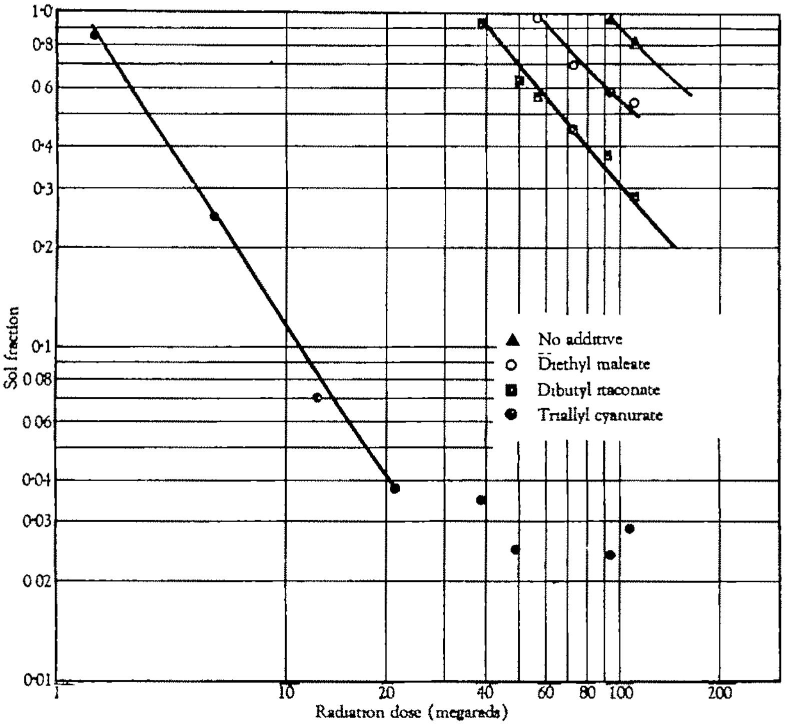

Fig. 1. Effect of unsaturated additives on polyvinyl acetate

that no gel fractions were obtained with or without the additive up to the highest dose used).

The superiority of triallyl cyanurate in enhancing cross-linking is believed to be due mainly to the presence of three unsaturated groups in the molecule. The much greater efficiency of triallyl esters compared with diallyl esters in polyvinyl chloride has already been noted[2].

Typical examples of the effect of these additives are shown in Figs. 1 and 2.

Fig. 1 shows the variation of sol fraction with dose for polyvinyl acetate alone and when mixed with the unsaturated compounds. The curves through the experimental values are drawn for degradation to cross-linking (β/α) ratios of 0·4 (polymer alone), 0·4 (polymer plus diethyl maleate), 0·2 (polymer plus dibutyl itaconate) and 0·0 (polymer plus triallyl cyanurate).

Fig. 2 similarly shows the variation of sol fraction with dose for polymethyl acrylate alone and when mixed with the additives. The curves are drawn for β/α ratios of 0·2 (polymer alone or mixed with diethyl maleate or dibutyl itaconate) and 0·0 (polymer plus triallyl cyanurate). While it is found that the addition of triallyl cyanurate to a polymer frequently gives a β/α ratio of 0·0 on irradiation, the other additives affected this ratio to a much smaller extent.

Poly*iso*butylene and polymethyl methacrylate are examples of polymers which have only a negligible tendency to develop cross-links under ionizing radiation. However, while mixtures of poly*iso*butylene with the additives became degraded, polymethyl methacrylate containing triallyl cyanurate gelled readily. This comparison suggests that the cross-linking of polymethyl methacrylate by triallyl cyanurate occurs through the ester groups. Comparison of energy yields for gas evolution and main-chain scission in polymethyl methacrylate indicates that each scission is accompanied by the decomposition of approximately one side-chain[3]. If it is assumed that addition of triallyl cyanurate results in each side-chain decomposition becoming converted to one cross-link, then the variation of sol fraction with dose would be expected to correspond with a β/α ratio of about 1. In fact, the variation of sol fraction with dose corresponds to β/α less than 0·4, so that it appears that the additive can interfere directly with main-chain scission unless, as has been tentatively suggested[3], main-chain scission arises as a direct result of side-chain scission in this polymer.

The results obtained with polypropylene are of interest in that the enhancement of cross-linking decreased with increase of the time between preparation of the sample and its irradiation. This may be due to the insolubility of the additive which immediately after compounding is present as a dispersion in the polymer, but on standing, diffuses to the surface, thus reducing the bulk concentration.

The frequent correspondence of the sol fraction–dose curves with the theoretical curves for a particular value of the β/α ratio suggests that graft polymerization of the additive is not the correct explanation of this enhancement of cross-linking. This conclusion is supported by the fact that one of the additives, diethyl maleate, has a marked reluctance to homo-

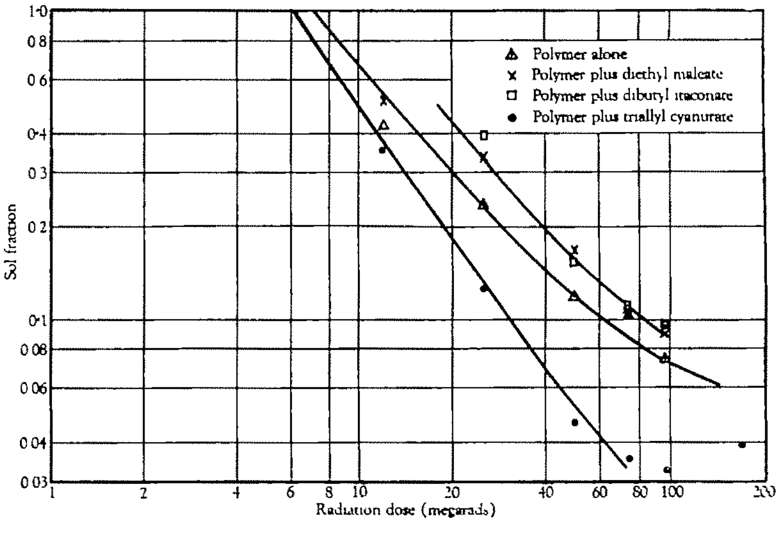

Fig. 2. Effect of unsaturated additives on polymethyl acrylate

polymerize and by the low rate at which such a polymerization would occur with the other additives at the concentration used and in such viscous media.

The additives, particularly triallyl cyanurate, seem to operate either by inhibiting chain scission in these polymers or by converting chain scission into a cross-linking reaction. It is possible that these additives may react with all the primary products of the action of ionizing radiation on the polymer. Examination of the effect of hydrolysis on the gel fraction of triallyl cyanurate cross-linked polymers may indicate the nature and extent of the interaction.

It is hoped to report the results of this investigation more fully at a later stage.

My thanks are due to Dr. R. M. Black for his advice and encouragement and to Dr. L. G. Brazier, director of research, British Insulated Callender's Cables, Ltd., for permission to publish this communication.

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[1] Black, R. M., and Lyons, B. J., *Proc. Roy. Soc.*, A, **253**, 322 (1959).

[2] Pinner, S. H., *Nature*, **183**, 1108 (1959).

[3] Bovey, F. A., "The Effects of Ionizing Radiation on Natural and Synthetic High Polymers" (Interscience Pub., New York, 1958).

Second Explosion Limit of Dried Carbon Monoxide – Oxygen Mixtures

The position of the second explosion limit of carefully dried mixtures of carbon monoxide and oxygen in quartz vessels has been determined. Both the heating and withdrawal methods were used in a static system. Carbon monoxide was prepared from sodium formate and concentrated sulphuric acid, and oxygen by heating potassium permanganate. After being passed through chemical desiccants and then through cold traps, the gases underwent a final drying operation immediately before use. This final drying consisted in allowing the gases (sometimes premixed) to stand in 'Pyrex' drying vessels surrounded by liquid oxygen for at least 12 hr.

Explosion limits for carbon monoxide – oxygen (1 : 2) mixtures in the pressure-range 40–250 mm. mercury lay between the temperatures 750° and 840° C., and are shown in Fig. 1 together with the published results of other workers. It will be seen that the present results lie at considerably higher temperatures than so far reported. Experiments were also made using gas mixtures which had not undergone the *final* drying procedure mentioned above. The limits were found to lie at a temperature some 100° C. lower. Gordon and Knipe[1] have shown that small traces of water vapour lower considerably the limit temperature. We suggest, therefore, that the higher limits reported here can be attributed to the use of drier gases than those employed in earlier work.

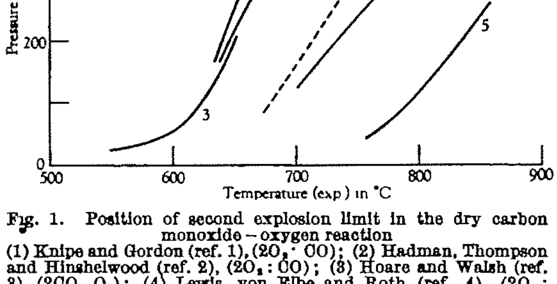

Fig. 1. Position of second explosion limit in the dry carbon monoxide – oxygen reaction
(1) Knipe and Gordon (ref. 1), $(2O_2 \cdot CO)$; (2) Hadman, Thompson and Hinshelwood (ref. 2), $(2O_2 : CO)$; (3) Hoare and Walsh (ref. 3), $(2CO \,.\, O_2)$; (4) Lewis, von Elbe and Roth (ref. 4), $(2O_2 : CO)$; (5) present work $(2O_2 \,.\, CO)$; (6) present work with the final drying process omitted

A detailed report of this and related work, including an account of the effects of mixture composition and of added inert gases, will be published elsewhere.

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[1] Gordon, A. S., and Knipe, R. H., *J. Phys. Chem.*, **59**, 1160 (1955).

[2] Hadman, G., Thompson, H. W., and Hinshelwood, C. N., *Proc. Roy. Soc.*, A, **138**, 297 (1932).

[3] Hoare, D. E., and Walsh, A. D., *Trans. Farad. Soc.*, **50**, 37 (1954).

[4] Lewis, B., von Elbe, G., and Roth, W., Fifth Symposium (International) of Combustion, 610 (1954).

New Approach to the Synthesis of O-Phosphoserine and Related Peptides

A general method of synthesizing phosphorylated amino-hydroxy acids and derived peptides, using diphenylphosphorylchloridate as the phosphorylating agent, was introduced by Riley *et al.*[1]. This method has been applied with rather satisfactory results in the synthesis of α-carbethoxy-L-lysyl-L-(O-phosphoryl)-serylglycine[2]; but further studies on the inhibitory spectrum of phosphoserine[3] towards proteolytic enzymes, and the effect of phosphatases[3] upon various substrates as well, stress the necessity of alternate synthetic methods.

When diphenyl groups from O-diphenylphosphoryl peptide derivatives containing other than aliphatic side-chain amino-acids are removed by catalytic hydrogenation in the presence of platinum catalyst, the likelihood of reduction of the side-chain rings is apparent. In this connexion it is of interest to note that O-diphenylphosphoryl esters have been believed up to the present to resist hydrogenolysis in the presence of palladium catalyst. Based on this assumption, a stepwise removal of protecting groups from carbobenzoxy-D,L-(O-diphenylphosphoryl)serine benzyl ester has been recently reported[4]. This consists, first, in the removal of the carbobenzoxy and benzyl groups by hydrolysis in the presence of palladium and then of the phenyl groups using platinum catalyst. Thus, the intermediate product D,L-(O-diphenylphosphoryl)serine[4] was described as a crystalline compound with melting point 129–130°.

Our experience, however, with hydrogenolysis of O-diphenylphosphoryl serine peptide derivatives in the presence of palladium has led us to a different conclusion. Special emphasis has been placed so far on the hydrolysis of carbobenzoxy-D,L-(O-diphenylphosphoryl)serine benzyl ester. When the latter was hydrogenated in the presence of a palladium catalyst and two equivalents of hydrogen were absorbed in about 1 hr., we were unable to isolate the reported product O-diphenylphosphoryl serine[4].

Surprisingly enough, the oily product obtained, which is soluble in water, was proved by paper chromatography to be a mixture of at least three components. The major product consisted of monophenylphosphorylserine[5], while two other unknown compounds were travelling with entirely different R_F value than that reported[4] for O-diphenylphosphorylserine.

Though the elucidation of this finding is still in progress, it is tempting to speculate that the emergence of the carboxyl- or amino-group of serine during the hydrolysis influences the elimination of a phenyl group via the formation of an intermediate six- or five-membered ring.

Unexpected difficulties were encountered also when diphenylphosphoryl chloridate was used to phosphorylate α-carbethoxy-(ε-carbobenzoxy)-L-lysylglycyl-L-serine benzyl ester. The latter peptide derivative was synthesized as follows: Tritylglycine[6] was coupled with serine benzyl ester by the carbodiimide method, and the thick oily product thus obtained was further detritylated to give glycyl-L-serine benzyl ester hydrochloride (I) in 75 per cent yield, melting point 173–175° (decomp., softens at 165–166°). Analysis: calc., $C_{12}H_{17}N_2O_4Cl$, N 9 70, Cl 12·28; found, N 9·50, Cl 11·90.

Compound I was coupled with α-carbethoxy-(ε-carbobenzoxy)-L-lysine[2] as previously described. In our experience and in spite of a previous statement[7], coupling in anhydrous solution leads to a considerable extent to the formation of the O-peptide isomer, which is difficult to remove.

The tripeptide derivative obtained, α-carbethoxy-(ε-carbobenzoxy)-L-lysylglycyl-L-serine benzyl ester (II), melted at 141–142° (sharp), yield 40–45 per cent, $[\alpha]^{23}_{D}$ 7·4 (c. 0·54 in glacial acetic acid). Analysis: calc., $C_{29}H_{38}N_4O_9$, C 59·36, H 6·52, N 9·7; found, C 59 45, H 6·72, N 9·53.

Compound II was phosphorylated with diphenylphosphorylchloridate in a similar manner to that described for its analogue, α-carbethoxy-(ε-carbobenzoxy)-L-lysyl-L-serylglycine benzyl ester[3]. Several attempts to obtain the product in a solid form have failed, probably owing to the presence of unphosphorylated peptide derivative which is difficult to separate by fractional precipitations. When a larger excess of chloride was used the result turned out to be less satisfactory. The oily product obtained, when hydrogenated and studied by paper chromatography, was found to give a mixture of several ninhydrin-positive spots. The possibility of some splitting of peptide bonds was naturally borne in mind[8].

In the course of our studies for improved methods for the synthesis of O-phosphorylated peptides, we have employed di-p-nitrobenzylphosphorylchloridate[9] as the phosphorylating agent.

Since benzyl groups are split off by catalytic hydrogenation with palladium catalyst, this method may be of wider application than that employing diphenylphosphorylchloridate.

Moreover, di-p-nitrobenzylphosphorylchloridate was expected to give readily crystallizing products as in the case of N-phosphoryl amino-acids and peptides[10]. Indeed, we have succeeded in preparing carbobenzoxy-D,L-(O-di-p-nitrobenzylphosphoro)-serine benzyl ester in crystalline form in 50–60 per cent yield. The product was melted at 107–108°, while the corresponding diphenylphosphoryl derivative has a considerably lower melting point (49–50°) and is difficult to crystallize. Analysis: calc., $C_{32}H_{30}N_3O_{12}P$, N 6·1, P 4·57; found, N 6·2, P 4·85.

Tetra-p-nitrobenzylpyrophosphate, readily prepared from di-p-nitrobenzylphosphate and carbodiimide in dimethylformamide, constitutes another route under investigation. So far we have succeeded in phosphorylating α-amino groups in the presence of tertiary base. Thus, N-di-p-nitrobenzylphosphoroglycine benzyl ester was prepared in about 50 per cent yield, melting point 111–112°. Analysis: calc., $C_{23}H_{22}N_3O_9P$; N 8·15, P 6·09; found, N 8·05, P 6·32.

The potentialities of di-p-nitrobenzylphosphorylchloridate and tetra-p-nitrobenzylpyrophosphate for the synthesis of O-phosphorylated peptides will be discussed elsewhere.

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[1] Riley, G., Turnbull, J., and Wilson, W., *J. Chem. Soc.*, 1373 (1957).

[2] Theodoropoulos, D., Bennich, H., Folsch, G., and Mellander, O., *Nature*, **184**, 187 (1959).

[3] Theodoropoulos, D., Bennich, H., and Mellander, O., *Nature*, **184**, 270 (1959).

[4] Folsch, G., and Mellander, O., *Acta Chem. Scand.*, 11, 1232 (1957).

[5] Bevan, T., Malkin, T., and Tiplady, J., *J. Chem. Soc.*, 3086 (1957).

[6] Zervas, L., and Theodoropoulos, D, *J. Amer. Chem. Soc.*, 78, 1359 (1956). Stelakatos, G., Theodoropoulos, D., and Zervas, L., *J. Amer. Chem. Soc.*, 81, 2884 (1959).

[7] Folsch, G., *Acta Chem. Scand.*, 12, 561 (1958).

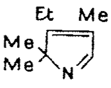
[8] Baer, E, Maurukas, J., and Clarke, D., *J. Biol. Chem.*, **228**, 181 (1957).

[9] Zervas, L., and Dilaris, I., *J. Amer. Chem. Soc.*, 77, 5354 (1955).

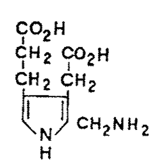
[10] Zervas, L., and Katsoyannis, P., *J. Amer. Chem. Soc.*, 77, 5351 (1955).

Formation of I- and III-Type Porphyrins by the Polymerization of Pyrroles

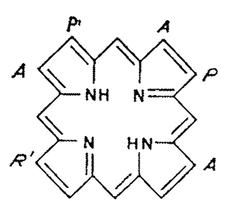
IN a recent paper[1] we discussed the alkylation of tri- and tetra-alkylpyrroles and referred to the 2 : 3 : 4-trialkylpyrroles which on alkylation could give either 2 : 3 : 4 : 5-tetra-alkylpyrroles or 2 : 2 : 3 : 4-tetra-alkyl-2(H)pyrroles (for example, I). The factors which decided the course of this reaction were delicately balanced, for whereas the methylation of the Grignard derivative of 2 : 3 : 4-trimethylpyrrole gave 2 : 3 : 4 : 5-tetramethylpyrrole almost exclusively, a similar methylation of 2 : 4-dimethyl-3-ethylpyrrole (cryptopyrrole) gave 3-ethyl-2 : 2 : 4-trimethyl-2(H)pyrrole (I) as the major product.



(I)



(II)

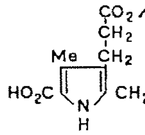


(III)

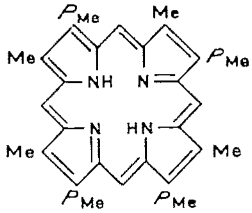
$A = \cdot CH_2CO_2H$
$P = \cdot CH_2CH_2 \cdot CO_2H$

This duality of reaction path has some interesting consequences, in particular in considering the biogenesis of natural porphyrins which are derived by the polymerization of porphobilinogen (II). Most of the common porphyrins belong to the so-called III series, for example, uroporphyrin III (III; $R = P$, $R' = A$). Although members of the I series, for example uroporphyrin I (III; $R = A$, $R' = P$), are also natural products, the II and IV series apparently do not occur naturally. In considering the mechanism by which the I- and more especially the III-type porphyrins might be formed from porphobilinogen it is clear that the initial reaction is the electrophilic alkylation of one molecule of porphobilinogen, a 2 : 3 : 4-trialkylpyrrole, by another porphobilinogen molecule. We have suggested[2] that, if this occurs at the 5-position, as in the methylation of 2 : 3 : 4-trimethylpyrrole, in a repeating process then uroporphyrin I will be the eventual product; if reaction occurs at the 2-position as in the methylation of cryptopyrrole, then uroporphyrin III results after a series of specified migrations. It was therefore of interest to ascertain how far this type of polymerization could be controlled to give porphyrins of the I or III series. Such reactions have considerable biochemical as well as chemical interest as certain porphyrias are known to be associated with the biological production of I- rather than III-type porphyrins. Cookson and Rimington[3] examined the polymerization of porphobilinogen under various conditions *in vitro* and found that, at 100°, uroporphyrin III was obtained rapidly and in high yield with no trace of the series I isomer. At 20°, the polymerization took several days and was dependent on pH; at pH 6·5, only the series III isomer was formed, but at pH > 10, equal amounts of the series I and III isomers were obtained. These results have been confirmed by Mauzerall and Granick[4].

We have now shown that these observations can be extended to the coproporphyrin series. Thus whereas the acid-catalysed polymerization of the pyrrole (IV; R = Me) had been shown[5] to yield coproporphyrin III tetramethyl ester (V), m.p. 151–154°, as the sole recognizable product, it has now been found that if the polymerization is effected by hot ethanolic ammonia, the product is mainly coproporphyrin I tetramethyl ester, m.p. 237–240°, containing some of the series III isomer.



(IV)

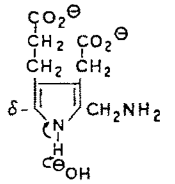


(V)

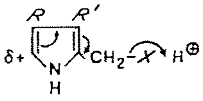
(P_{Me} = ·CH₂ CH₂·CO₂Me)

Likewise, polymerization of the corresponding dicarboxylic acid (IV; R = H) in ethylene glycol solution in presence of sodium carbonate, gave a product which, after esterification by methanolic hydrogen chloride, could be resolved by chromatography into fractions which were mainly coproporphyrin I tetramethyl ester on one hand and mainly the series III isomer on the other. These identifications were made on the basis of melting points of the methyl esters and chromatographic examination of the free porphyrins in the system 2 : 6-lutidine–water, and we are very grateful to Prof. C. Rimington and Mrs. A. Latter, who carried out these determinations.

The course of these *in vitro* polymerizations is therefore controlled by pH, a high pH leading to the formation of a mixture of I- and III-type porphyrins and a low pH leading only to the III-type. From the mechanistic point of view, the alkaline polymerization is regarded as involving a loosening of the pyrrole N–H bond by the alkali, leading to an increased electron density in the pyrrole ring (as in VI). This enhances the ease of electrophilic substitution at the vacant 5-position and leads eventually to I-type porphyrins. Ionization of the side-chain carboxyl groups in porphobilinogen will doubtless accentuate this effect, but the parallel results with the pyrrole esters (for example, IV) show that the free acid groups are not essential.



(VI)



(VII)

In acidic media, a pyrrole of type (VII) will tend to ionize in the manner shown (X = OAc, Br or $\overset{+}{\mathrm{N}}\mathrm{H}_3$ in the protonated form of porphobilinogen), which causes a deactivation of the ring towards electrophilic attack of the 5-position and thus favours reaction at position 2-. This substitution pattern leads to the formation of III-type porphyrins. It is not yet clear how far these ideas can be extended to the *in vivo* polymerizations of porphobilinogen to uroporphyrins I and III, but it would appear probable that certain aspects of the reaction mechanisms are common to both *in vivo* and *in vitro* porphyrin syntheses. These views were communicated to Prof. A. Neuberger, who included them in his Section Lecture to the Seventeenth International Congress of Pure and Applied Chemistry at Munich, 1959.

In recent papers, Bogorad[7] describes the isolation of an enzyme, porphobilinogen deaminase, which causes the polymerization of porphobilinogen to uroporphyrinogen I (cf. alkaline catalysis), but in the presence of a second enzyme, uroporphyrinogen isomerase, the product was uroporphyrinogen III (cf. acid catalysis). It is possible that some chemical interaction occurs between the two enzymes which causes a modification of the essential functional groupings, and it may not be necessary to postulate, as Bogorad does, a substrate other then porphobilinogen for the isomerase.

In order to meet Bogorad's suggestions, Wittenberg has recently[8] published a mechanism for porphobilinogen polymerization which involves an octapyrrolic intermediate with successive dehydrogenation and hydrogenation steps. Apart from the improbability that a 24-membered macrocylic ring would be formed in such a facile reaction, the theory does not offer a convincing explanation of the exclusive production of III-type porphyrins under conditions involving acid catalysis. Moreover, it postulates for the formation of the tripyrromethane portions of the large macrocycle a type of condensation which has not been observed in the laboratory.

Wittenberg also refers to the problem of the biogenesis of vitamin B_{12} which, like the porphyrins, has porphobilinogen as a pyrrolic intermediate. He makes the point that if the formation of the linkage between rings A and D is the initial step in the polymerization, as we suggest, this does not accord with the view we have previously advanced[9], that the final ring closure in the formation of vitamin B_{12} is the formation of the direct link between rings A and D. These apparently contradictory views can be reconciled if an additional rearrangement of uroporphyrinogen III is involved. The nature of this reaction is unknown but a pinacol rearrangement of the type visualized by Woodward[10] to explain the exocyclic C_1 substituent in magnamycin is a possibility and is being examined in our laboratory.

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[1] Booth, Johnson, Markham and Price, *J. Chem. Soc.*, 1537 (1959).

[2] Bullock, Johnson, Markham and Shaw, *J. Chem. Soc.*, 1430 (1958).

[3] Cookson and Rimington, *Biochem. J.*, **57**, 476 (1954).

[4] Mauzerall and Granick, Fourth Intern. Congr. Biochem., Vienna, 4 (Pergamon Press, London, 1958).

[5] Johnson, Kay, Markham, Price and Shaw, *J. Chem. Soc.*, 3416 (1959).

[6] Booij and Rimington, *Biochem. J.*, **65**, 3,P, 4,P (1957). Granick and Mauzerall, *J. Biol. Chem.*, **232**, 1119, 1141 (1958). Bogorad, *ibid.*, **233**, 501, 500, 516 (1958)

[7] Bogorad, *J. Biol. Chem.*, **233**, 501, 510 (1958).

[8] Wittenberg, *Nature*, **184**, 876 (1959).

[9] Bonnett, Cannon, Johnson, Sutherland, Todd and Smith, *Nature*, **176**, 328 (1955).

[10] Woodward, *Angew. Chem*, **69**, 50 (1957).

BIOCHEMISTRY

Free Amino-acids of Some Fungi

THE nature and amounts of free amino-acids in higher plants are becoming relatively well known since the advent of paper chromatography. However, few investigations of amino-acids in the Cryptogams have been made. Fowden[1] has studied the amino-acids of certain algae, Mansford and Raper[2] those of *Mucor mucedo*, Fluck and Richle[3] those of *Fusarium lycopersici*. The yeasts, *Aspergillus*, *Penicillium* and *Neurospora* species[4], have been extensively studied but the rest of the fungi remain largely unexamined.

Robbins[5] has classified fungi physiologically according to their ability to utilize various forms of nitrogen: Group I, Molecular nitrogen, NO_3^-, NH_4^+ and organic nitrogen; Group II, NO_3^-, NH_4^+ and organic nitrogen; Group III, NH_4^+ and organic nitrogen; Group IV, organic nitrogen.

It appeared desirable to study the free amino-acids of a taxonomic range of fungi which included most of these physiological groups. The species chosen were as follows: Phycomycetes—*Pythium ultimum* Trow. (Group II); *Phytophthora cactorum* Leb. and Cohn (Schroet.) (Group IV); *Phycomyces nitens* + (Agardh) Kunze and Schmidt (Group IV); *Thamnidium elegans* Link (Group III). Ascomycetes and Fungi Imperfecti—*Chromocrea spinulosa* (Fuckel) Petch (Group II); *Fusarium culmorum* (W.G.Sm.) Sacc. (Group II); *Fusarium javanicum* Koorders (Group II). Basidiomycetes—*Stereum purpureum* Pers. ex Fries (Group II).

Pythium ultimum was cultured on liquid Czapek–Dox medium with nitrate as the nitrogen source and sucrose as the carbon source. The others were grown on a liquid medium in which asparagine (0·3 per cent) was the nitrogen source and glucose (2 per cent) the carbon source. Minerals and vitamins included potassium dihydrogen phosphate (0·1 per cent), magnesium sulphate and potassium chloride (0·05 per cent) and trace amounts of Ca^{++}, Fe^{++}, Zn^{++}, Cu^{++}, Mn^{++}, MoO_4^- and thiamine. All fungi were grown at 25° C. as stationary cultures in 250 ml. Erlenmeyer flasks containing 100 ml. liquid medium.

After varying intervals of time depending on the species growth-rate, fully developed mycelial pads still in growth were separated from the medium and were washed once with distilled water. The amino-acids were then extracted by blending the hyphæ with 87 per cent alcohol at room temperature. The extracts were concentrated and then examined for amino-acids by paper chromatography.

A summary of the free amino-acids found in the fungal hyphæ is presented in Table 1.

Several amino-acids were found in all the fungi studied. Fowden[1] found a similar pattern in three species of algae. The amino-acid, γ-amino butyric, has been found in a wide range of plants and it is interesting to record its presence in seven of the eight fungi examined. It is not a constituent of protein hydrolysates, nor is β-alanine, which was found in four of the fungi. Large amounts of proline

Table 1. FREE AMINO-ACIDS PRESENT IN THE HYPHAL EXTRACT OF CERTAIN FUNGI

	Pythium ultimum (on Dox)	*Phytophthora cactorum*	*Phycomyces nitens* +	*Thamnidium elegans*	*Chromocrea spinulosa*	*Fusarium culmorum*	*Fusarium javanicum*	*Stereum purpureum*
Aspartic acid	+ +	+ +	+ + +	+ + +	+ +	+ + +	+ + +	+ + +
Glutamic acid	+ +	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +
α-Alanine	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +
β-Alanine	—	+ +	+	+	—	—	+	—
γ-Amino butyric acid	+ +	+ + +	+ +	+ +	+ + +	+ +	+	—
Serine	+	+ +	+ +	+	+ +	+ + +	+ +	+ +
Glycine	+ +	+ +	+ +	+	+ +	+ +	+ +	+
Threonine	+ +	+ +	+ +	—	+	+	+ +	+
Proline	—	+ + +	—	+ +	—	—	—	—
Tyrosine	—	+	+	—	+	—	+	—
Arginine	+ + +	+ + +	+ + +	+	+ +	+ +	+ +	+ +
Histidine	+ +	+ +	+ +	+	+ +	+ +	+ +	+ +
Lysine	+ +	+ +	—	—	+ +	—	+ +	+ +
Phenylalanine	—	—	+	+	+	—	—	—
Leucine and/or *iso*-leucine	+ +	+ +	+ +	+	+ +	+ +	+ +	+
Methionine and/or valine	+ +	+ +	+ +	+	+ + +	+ +	+ +	+
Ornithine	—	+	—	—	—	+	—	+
Cysteic acid	+ +	+	—	+	+ +	+ +	+	—
Asparagine	+ +	+ + +	+ + +	+ + +	+ + +	+ +	+ + +	+ + +
Glutamine	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +
Initial *p*H	3·7	4 2	4·2	4·2	4·2	4·2	4·2	4·2
Final *p*H	5·5	4 7	6·7	3 9	5·1	7·0	6 9	6·5
Incubation time (days)	10	29	16	26	14	14	24	19

+ + +, Strong ninhydrin colour, + +, moderate colour +, weak colour; —, not detected.

were found in *Phytophthora cactorum* and its presence was noted in *Thamnidium elegans*. Tyrosine was present in small amount in several, but tryptophan was not detected in any of the fungi. Glutamine appeared to be present in larger amounts than asparagine. Four unidentified compounds were found in some of the fungi. These corresponded on chromatograms to the positions of α-amino adipic acid, 3 : 4-dihydroxyphenylalanine, ethanolamine and taurine, but identification must await further work.

Under these experimental conditions, there do not appear to be any substantial differences between fungi belonging to different taxonomic and physiological groups. The number and relative amounts of free amino-acids present in fungi show an overall similarity to those in higher plants.

The assistance of Dr. E. G. Bollard is gratefully acknowledged.

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[1] Fowden, L., *Nature*, **167**, 1030 (1951).
[2] Mansford, K., and Raper, R., *Nature*, **174**, 314 (1954).
[3] Fluck, V., and Richle, K. H., *Phytopath. Z.*, **24**, 455 (1955)
[4] Foster, J. W., "Chemical Activities of Fungi" (1949).
[5] Robbins, W. J., *Amer. J. Bot.*, **24**, 243 (1937).

Specificity *in vitro* of a Phenoloxidase System from *Periplaneta americana* (L.)

ENZYMES capable of oxidizing mono- and diphenols are widely distributed in insects; some of these compounds are involved in the biosynthesis of exoskeletal substances, notably sclerotin[1]. While several *ortho*- and *para*-diphenols have been extracted from cuticle, relatively little is known about the corresponding synthetic enzymes. Recent work in connexion with the reaction of phenolic derivatives with proteins ('tanning reaction') has been reviewed by Hackman[2], Mason[3] and Dennell[4]. The present results form part of an investigation of the intermediary metabolism of phenolic substrates involved in sclerotin formation in *Periplaneta*.

It has been already shown that the sclerotin walls of the egg capsule (ootheca) are formed by the intermingling of the secretions of the left and right colleterial glands[1]. As a result of this, protocatechuic acid[5] (3 : 4-dihydroxybenzoic acid) is liberated from its 4-O-β-D-glucoside (secreted by the left gland) by the action of a β-glucosidase (from the right gland)[6]. Some evidence[1] suggests that the protocatechuic acid is oxidized enzymically by a secretion of the left gland[7], afterwards to link with structural proteins to form sclerotin.

It has now been confirmed that the oxidative enzymes are present in the lumen of the left gland and are released in the form of a highly turbid suspension when the dissected left gland is allowed to stand for a few minutes in distilled water. The preparation can be freed from cellular debris by centrifugation at 0° C. and from endogenous substrates by dialysis ('Cellophane') against 0·01 *M* sodium chloride. Subsequent salt precipitation gave fractions of very high enzymic activity between 0·1 *M* and 0·2 *M* sodium chloride (final concentration).

The activity of the dialysed enzyme preparations was investigated in 0·1 *M* sodium phosphate buffer (*p*H 6·8) with a variety of phenolic compounds (each in a final concentration of 0·002 *M*) using Warburg manometers (5–6 ml.) at 37° in an atmosphere of oxygen. The results are shown in Table 1.

Table 1. SUBSTRATES FOR THE PHENOLOXIDASE SYSTEM FROM LEFT COLLETERIAL GLANDS OF *Periplaneta*

Active	Inactive
Protocatechuic acid	DL-3 4-Dihydroxyphenylalanine
3 : 4-Dihydroxybenzoic acid	L-Tyrosine (*para*)
3 : 4-Dihydroxypropionic acid	L-Tyrosine (*ortho*)
3 : 4-Dihydroxybenzaldehyde	*p*-Hydroxybenzoic acid
Catechol	DL-3 : 4-Dihydroxyphenylethylamine (dopamine)
Hydroquinone	Resorcinol
p-Phenylenediamine	

The enzyme promoted rapid oxidation of a number of *o*- and *p*-diphenols with the notable exception of 3 : 4-dihydroxyphenylalanine and the corresponding amine. L-Tyrosine (either *ortho* or *para*) or other monophenols were not oxidized.

Thus, in the sense of Keilin and Mann[8] and Dawson and Tarpley[9], this enzyme can be regarded as a laccase and in no sense a tyrosinase.

The enzyme activity is abolished by being heated above 70° and by 0·002 *M* potassium cyanide. It is unaffected by 4-chlororesorcinol and diethyldithiocarbamate. Preliminary investigations indicate that the enzyme is a protein–copper complex.

It is emphasized that the enzyme preparation has not yet been obtained in a soluble form but that it occurs in the gland in a finely divided state exhibiting a strong Tyndall effect. This is supported by electron microscopy[10].

It has not yet been found possible to separate the enzyme from the protein-precursor of sclerotin, of which indeed the enzyme may be a part.

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[1] Pryor, M. G. M., *Proc. Roy. Soc.*, B, **128**, 378 (1940).
[2] Hackman, R. H., Proc. Fourth Internat. Cong. Biochem., Vienna, **12**, 48 (1958).
[3] Mason, H. S., "Adv. in Enzymol.", **16**, 105 (1955).
[4] Dennell, R., *Biol. Rev.*, **33**, 178 (1958).
[5] Pryor, M. G. M., Russell, P. B., and Todd, A. R., *Biochem. J.*, **40**, 627 (1946).
[6] Brunet, P. C. J., and Kent, P. W., *Proc. Roy. Soc.*, B, **144**, 259 (1955).
[7] Brunet, P. C. J., *Quart. J. Micro. Sci.*, **93**, 47 (1952).
[8] Keilin, D., and Mann, T., *Nature*, **143**, 23 (1939).
[9] Dawson, C. R., and Tarpley, W. B., in "The Enzymes", ed. by Sumner, J. B., and Myrbäck, K., **2**, Pt. 1, 454 (1951).
[10] Mercer, E. H., and Brunet, P. C. J., *J. Biophys. Biochem. Cytol.*, **5**, 257 (1959)

Infra-red Spectra of Capsular Polysaccharides from Thirteen Strains of *Cryptococcus neoformans*

STUDIES of infra-red absorption spectra of immunopolysaccharides have received little attention until recently. Thus, Levine, Stevenson and Kabler[1] were able to show that the serologically reactive and type specific pneumococcal polysaccharides have distinctive spectra by which they can be identified. Later, Watson, Marinetti and Scherp[2] working with the specific hapten of group *C* meningococcus were successful in assigning a provisional structure for the hapten by the aid of infra-red spectral analysis. Inasmuch as purified cryptococcal polysaccharides have shown serological reactivity by precipitation

Table 1. INFRA-RED SPECTRAL DATA OF *C. neoformans* POLYSACCHARIDES

Sample*	Absorbance						Source
	5·77μ	6·24μ	6·85μ†	7·07μ	7·27μ†	8·0–8·2μ	
BRI-C	0·275	0·241	0·242	0·271	0·254	0·334	Human *CSF*‡
3723-*C*	0·358	0·410	0·405	0·454	0·418	0·487	Bovine mastitis
RE-C	0·392	0·425	0·425	0·463	0·446	0·512	Human *CSF*
2700-*C*	0·472	0·492	0·495	0·541	0·509	0·604	Pigeon environs
3717-*C*	0·449	0·455	0·452	0·505	0·468	0·561	Soil
3727-*C*	0·380	0·496	0·445	0·580	0·495	0·525	Bovine mastitis
2400-*C*	0·498	0·535	0·585	0·640	0·598	0·649	Human bone
2600-*C*	0·234	0·283	0·435	0·330	0·354	0·344	Pigeon environs
MY-C	0·495	0·488	0·438	0·560	0·526	0·621	Human *CSF*
BRI-P	0·425	0·421	0·516	0·470	0·519	0·501	Human *CSF*
3723-*P*	0·310	0·362	0·553	0·399	0·493	0·402	Bovine mastitis
DU-P	0·265	0·275	0·364	0·310	0·354	0·332	Human bone
TRE-P	0·349	0·365	0·505	0·410	0·486	0·435	Human *CSF*
HU-P	0·302	0·323	0·445	0·374	0·426	0·402	Human *CSF*
BYR-P	0·330	0·330	0·625	0·380	0·576	0·370	Human *CSF*

* Sample: *C*, crude preparation; *P*, pure (capable of stimulating antibody formation in mice).

† Corrected for 'Nujol' absorption.

‡ *CSF*, Cerebrospinal fluid.

with specific hyperimmune rabbit serum[3,4] in addition to stimulating antibody formation in mice[5], it was deemed desirable to investigate their infra-red spectra and to compare these with the spectra of crude preparations.

Samples of the polysaccharides from pathogenic *Cryptococcus neoformans* strains were purified by the methods of Gadebusch[5], triturated in repurified commercial paraffin oil ('Nujol'), and spectra obtained from a paste thickness of approximately 0·01 mm. by means of a Perkin–Elmer double-beam recording spectrophotometer. In all cases the thinnest film possible was employed to bring out detail between 8·5 and 15μ. Crude polysaccharides were so designated because they had not been specifically deproteinized by the methods of Sevag[6].

In spite of the diverse origin of the cryptococcal polysaccharides (Table 1), the spectra were so strikingly similar that identification seems extremely difficult, if not impossible. Purification of these substances, while producing definite immunological differences[5], did not alter their infra-red spectra beyond a somewhat greater resolution of existing peaks. These spectral relationships have been borne out by analytical data which have shown similar values for carbon, hydrogen, oxygen, reducing sugar, uronic acid and acetyl groups (unpublished work). All these polysaccharides contain negligible amounts of nitrogen and phosphorus.

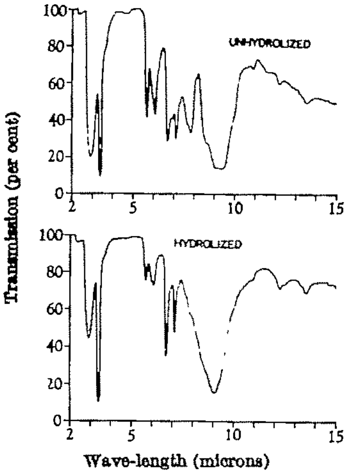

Fig. 1. Infra-red spectra of hydrolysed and unhydrolysed purified polysaccharide from *C. neoformans* strain *BRI*

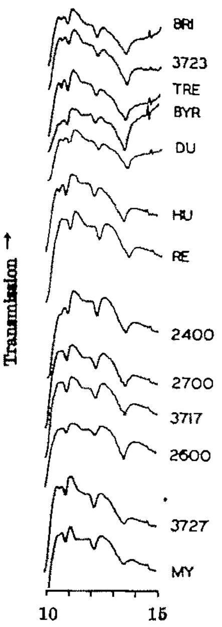

Fig. 2. Infra-red spectra of *C. neoformans* polysaccharides. The upper six spectra were from purified samples and the remainder were from crude preparations

As a result, it seems quite unlikely that capsular type identification as performed serologically by Evans[7] could be accomplished by a study of the infra-red spectra of their polysaccharides. In this respect at least spectral investigations of pneumococcal polysaccharides have been more encouraging[8].

Despite partial interference by 'Nujol' absorption (3·43μ, 6·85μ, 7·27μ), a few preliminary remarks may be made regarding the infra-red spectra of *C. neoformans* polysaccharides: (1) The source for the moderately strong carbonyl band at 5·77μ is probably the ester linkage and more specifically O-acetyl. Acids absorbing at this wave-length have been eliminated as a source, since precipitation was not carried out at low *p*H. In addition, the presence of the frequently accompanying 8·0–8·2μ band lends further confirmation to the ester origin. (2) Bands at 6·20 and 7·10μ are characteristic of the carboxylate ion which in turn may be derived from uronic acids. Since these polysaccharides all contain a uronate moiety, the bands at 6·24 and 7·07μ may well have been displaced somewhat. (3) Although bands at 6·85 and 7·27μ are partially attributable to 'Nujol' absorption, the intensity of absorption indicates the presence of another structural component, probably the —CH_3 group. (4) Carefully controlled hydrolysis of a water solution of *C. neoformans* strain *BRI* purified polysaccharide in the presence of sodium hydroxide produced a substantial diminution in the absorption spectra at 5·77, 6·24, 7·07, and 7·27μ (Fig. 1). The band at 8·0–8·2μ now appears only as a shoulder on the larger, broader band between 9 and 10μ. These changes have been interpreted as a combination of ester and uronate hydrolysis. The moderate decrease in absorption of the 7·27μ band indicates partial loss of this terminal —CH_3 group. (5) Finally, in the region above 8·5μ, the bands at 11·15, 12·50 and 13·85μ (Fig. 2) appear to be characteristic for both crude and purified cryptococcal polysaccharides.

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[1] Levine, S., Stevenson, H. J. R., and Kabler, P. W., *Arch. Biochem.*, 45, 65 (1953).

[2] Watson, R. G., Marinetti, G. V., and Scherp, H. W., *J. Immunol.*, 81, 337 (1958).

[3] Neill, J. M., Castillo, G. G., Smith, R. H., and Kapros, C. E., *J. Exp. Med.*, 89, 93 (1949).

[4] Evans, E. E., and Kessel, J. F., *J. Immunol.*, 67, 109 (1951).

[5] Gadebusch, H. H., *J. Infect. Dis.*, 102, 219 (1958).

[6] Sevag, M. G., Lackman, D. B., and Smolens, J., *J. Biol. Chem.*, 124, 425 (1938)

[7] Evans, E. E., *Proc. Soc. Exp. Biol. Med.*, 71, 644 (1949).

[8] Stevenson, H. J. R., and Levine, S., *Science*, 116, 705 (1952).

Carnitine in Lipid Metabolism

THE finding by Fraenkel and Friedman[1] that carnitine (4-trimethyl amino 3 hydroxy butyric acid) is an essential growth factor for certain organisms (vitamin B_T) and its availability by synthesis have renewed interest in the problem of its biochemical function.

Recent reports suggest that carnitine is implicated in lipid metabolism. Schaepdryver and Vleeschhouwer[2] showed that it increased the fat content of lymph; Fritz and McEwan[3] postulated a role in fat transport; and it has been reported present in vagotonin[4] and lipocaic[5]. Binon and Deltour[6] reported that, contrary to the belief that the nitrogenous base of dog serum phospholipid was all choline, one-third was actually carnitine.

The chemical similarity of carnitine and choline suggests that carnitine might possess anti-lipotropic activity. This possibility was examined by the 6-day mouse test of Welch and Welch[7]. Seventy young male mice were fed on (*a*) high fat, negative control diet; (*b*) positive controls containing 0·5 per cent choline; (*c*) test diet containing 0·5 per cent carnitine; and (*d*) test diet containing 1·0 per cent carnitine. The results (Table 1) show that carnitine had no effect on the fat content of the liver. This result substantiates the finding of Fritz[8] on rats.

As a further examination of the role of carnitine in lipid metabolism the finding of Binon and Deltour[6] was investigated. These authors analysed the total lipid fraction from dog serum by differential reineckate precipitation[9]. The precipitate at *p*H 10 was assumed to be choline, and that at *p*H 2, carnitine. The carnitine was not further identified.

Table 1. TOTAL LIPID CONTENT OF MOUSE LIVERS (FOUR ANIMALS GROUPED IN EACH DETERMINATION)

Group	High diet fat. Negative controls (choline + carnitine absent) (gm./100 gm.)	Plus 0 5 per cent choline. Positive controls (gm./100 gm.)	Plus 0·5 per cent carnitine (gm./100 gm.)	Plus 1·0 per cent carnitine (gm./100 gm.)
Total animals used	20	20	20	10 (2 per group)
1	54 2	10 9	58·6	58·5
2	55 4	12·4	55·0	57 2
3	52·2	19·9	55 4	47 5
4	59·0	11·2	51·8	47·1
5	60 7	22 5	53·0	62·1

The lipids from 300 ml. of foxhound serum were extracted with ethanol/ether and hydrolysed by boiling with methanolic hydrochloric acid (following the procedure of Binon and Deltour). The two fractions obtained by differential reineckate precipitation at *p*H 10 (voluminous) and *p*H 2 (trace) were examined by paper chromatography. They were run in the presence of silver nitrate[10] in ethanol/ammonium hydroxide/water (95 : 5 : 5) and in a second experiment in *iso*propanol/ammonium hydroxide/water (20 : 1 : 2). The spots were located with iodine and with potassium bismuth iodide. Choline was the only quaternary ammonium compound detected in both reineckate fractions. Direct chromatography of the phospholipid hydrolysate, without reineckate treatment, also failed to reveal the presence of carnitine.

In addition to the serum investigation the phospholipid fraction of beef muscle was examined. The fraction was obtained by petroleum ether/ethanol extraction followed by acetone precipitation[11]. The hydrolysed material showed no reineckate precipitate under acid conditions, and chromatography revealed choline as the only quaternary ammonium compound.

It thus appears that carnitine is not present in phospholipids of blood or muscle, and that it does not play a part in transmethylation reactions that lead to the formation of choline or betaine.

A detailed account of this work and further investigations into the role of carnitine will be published elsewhere.

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[1] Fraenkel, G., and Friedman, S., "Vitamins and Hormones", 15 (1957).

[2] Schaepdryver, A. F. de, and Vleeschhouwer, G. R. de, *Arch. Intern. Pharmacodynamie*, 113, 15 (1957).

[3] Fritz, I. B., and McEwan, B., *Science*, 129, 334 (1959).

[4] Binon, F., and Deltour, G., *Experientia*, 12, 357 (1956).

[5] Binon, F., and Deltour, G., *C.R. Soc. Biol.*, 149, 932 (1955).

[6] Binon, F., and Deltour, G., *Rev. fermentations et indus. aliment.*, 11, 14 (1956).

[7] Welch, M. S., and Welch, A. DeM., *Proc. Soc. Exp. Biol. Med.*, 39, 5 (1938).

[8] Fritz, I. B., *Amer. J. Physiol.*, 190, 449 (1957).

[9] Binon, F., *Voeding*, 16, 781 (1955).

[10] Bregoff, H. M., Roberts, E., and Delwiche, C. C., *J. Biol. Chem.*, 205, 565 (1953).

[11] Hardman, E. E., and Crombie, W. M. L., *J. Exp. Bot.*, 9, 239 (1958).

17-Oxosteroids of Pregnant Ewes' Urine

IN view of the increasing use of steroid hormones and stilbene derivatives in agriculture and agricultural research, it was felt that it was necessary to investigate steroid excretion by the ewe.

The isolation of 5β-pregnane-3α : 20α-diol from the urine of the pregnant ewe was recently reported by Robertson and Coulson[1]. As a continuation of this work the neutral ketonic fraction of pregnant ewes' urine was investigated.

22·5 l. of urine representing twenty 24-hr. collections from two ewes in the fourth month of pregnancy, were acid hydrolysed and extracted in the usual way. The neutral extract was separated into ketonic and non-ketonic fractions by the method of Dorfman[2].

The ketonic fraction of 100 mgm. was then chromatographed on an alumina column but no crystalline fractions were obtained.

The residues from the column were bulked into four main fractions (*A–D*), and 1/100 aliquots from each of these fractions were then chromatographed on paper in each of the following systems: (1) 100–120 petroleum ether–propylene glycol, (2) Bush's system *A*, (3) toluene–propylene glycol.

A series of known steroids were also run to provide standards and the chromatograms were sprayed with a modified Zimmerman reagent to locate 17-oxosteroid-like material. These results are summarized in Table 1.

Table 1. CHROMATOGRAPHY OF THE NEUTRAL KETONIC FRACTION OF PREGNANT EWES' URINE

Fraction	Substance†	Approximate quantity present‡	R_F value* in system		
			Petroleum ether–propylene glycol	Bush *A*	Toluene–propylene glycol
		(μgm.)			
A	*A*(1)	5	0·2	0·23	0·6
	A(2)	10–20	0·58	0·83	1·0
B	*B*(1)	10	0·2	0·23	not resolved
	B(2)	10	0·58	0·83	1·0
	B(3)	5	not resolved	0·68	not resolved
C	*C*(1)	5	not resolved	not resolved	0·15
D	No 17-oxosteroids	—	—	—	—
	Androsterone		1·0	1·0	1·0
	Aetiocholanolone		0·6	0·85	1·0
	Dehydro*epi*androsterone		0·5	0·69	1·0
	$\Delta^{1,4}$ Androstadiene-3 : 17-dione		0·55	0·55	1·3
	Δ^{4} Androstene-3 : 17-dione		1·25	0·86	1·3

* The rates of flow are expressed relative to androsterone.

† Only those materials which gave purple colour reactions similar to 17-oxosteroids are recorded.

‡ The approximate quantity of material present was deduced by comparison of the spot produced with one produced by 20 μgm. of steroid, taking into account the fact that the lower limit of sensitivity of the reagent was about 2·5 μgm. of steroid per spot.

Material *A*(1) and *B*(1) were apparently the same substance but could not be identified from the standards. This material like *C*(1) was of low R_F value and was therefore of a more polar nature than the standards.

From a comparison of R_F values substances *A*(2) and *B*(2) were apparently identical with aetiocholanolone.

Substance *B*(3) which was only resolved from *B*(2) in the Bush system and was therefore of very similar R_F value to *B*(2) in the other systems was tentatively identified as dehydro*epi*androsterone.

As 1/100 aliquots were run on paper the total 17-oxosteroid content of the urine was probably of the order of 5 mgm., which correspond to an excretion-rate of 0·25 mgm. per sheep per 24 hr. This appears to be made up of 2 mgm. (100 μgm. per sheep per 24 hr.) of aetiocholanolone, 500 μgm. (25 μgm. per sheep per 24 hr.) of dehydro*epi*androsterone and 2·5 mgm. (125 μgm. per sheep per 24 hr.) of at least two other unidentified steroids.

Thus the 17-oxosteroid excretion in pregnant ewes' urine, like the pregnanediol excretion, is very small by comparison with human urinary excretion.

As a result of the small quantity of 17-oxosteroids present and the large quantities of other material encountered it was concluded that the accurate quantitative estimation of 17-oxosteroids in sheep's urine could not be carried out by the usual methods.

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[1] Robertson, H. A., and Coulson, W. F., *Nature*, 182, 1512 (1958).

[2] Dorfman, R. I., Cook, J. W., and Hamilton, J. B., *J. Biol. Chem.*, 130, 285 (1939).

Isolation of 17-*Epi*œstriol from the Urine of Pregnant Women

THE isolation of 16α-hydroxyœstrone in 1957 by Marrian, Watson and Panattoni[1] led these authors to suggest that this compound might be the metabolic intermediate in the 'hydration' of œstrone to œstriol. In the same year, Brown and Marrian[2] reported that about 40 per cent of 16α-hydroxyœstrone when administered to human subjects was afterwards excreted in the urine as œstriol. Breuer, Nocke and Knuppen[3] found after incubation of 16α-hydroxyœstrone with slices of normal human liver in addition to œstriol a second metabolite which was identified as 17-*epi*œstriol (œstra-1,3,5-triene-3,16α,17α-triol). In view of these findings it seemed reasonable to assume that 17-*epi*œstriol might be a normally occurring metabolite of the œstrogens, and therefore, an attempt has been made to isolate 17-*epi*œstriol from urine during pregnancy.

Late-pregnancy urine was hydrolysed with concentrated hydrochloric acid, extracted with ether and the extract washed with an aqueous solution of sodium bicarbonate and water. The 'polar' fraction containing œstriol and its epimers was recovered by water extraction and then further purified by saponification. In order to separate the *cis*-glycols (16-*epi*œstriol and possibly 17-*epi*œstriol) from the *trans*-glycols (œstriol and 16,17-*epi*œstriol) the material was treated with anhydrous acetone containing 1 per cent hydrochloric acid. The acetonides were extracted with chloroform and hydrolysed with 0·5 *N* phosphoric acid. The fraction thus obtained was chromatographed in small portions on a large number of sheets of paper using the system formamide/chloroform. The areas corresponding to the position of 17-*epi*œstriol were eluted with methanol, the eluates combined and rechromatographed in the same system for 6 hr. After spraying the paper with Folin and Ciocalteu's reagent a blue spot was detected which had the same mobility (1·8 cm./hr.) as authentic 17-*epi*œstriol. In a second experiment, the areas expected to contain 17-*epi*œstriol were eluted, the eluates evaporated and subjected to sublimation under normal pressure. Crystals were obtained which showed the same melting point (233–235°) as authentic 17-*epi*œstriol. The material had the same absorption curve as 17-*epi*œstriol in the ultra-violet region (maximum at 279 mμ), with the Kober-reaction (maximum at 515 mμ) and with the sulphuric acid/water reaction (maxima at 286 and 456 mμ). The mobility of the fast black salt potassium derivative (0·33 cm./hr.) was identical with that of 17-*epi*œstriol when chromatographed in the system toluene/light petroleum/ethanol/water (200 : 100 : 30 : 70).

In view of the evidence presented here it is concluded that in addition to œstriol[4], 16-*epi*œstriol[5] and 16,17-*epi*œstriol[6], the fourth possible isomer, 17-*epi*œstriol, is present in human pregnancy urine.

As compared with œstriol, however, its concentration is very small. In a pooled late-pregnancy urine, the yield of 17-*epi*œstriol as determined by the Kober reaction was approx. 10 μgm./l., whereas the concentration of œstriol amounted to 15 mgm./l.

Full experimental details will be published elsewhere. I am grateful to Dr. R. Knuppen for preparing 17-*epi*œstriol, to Dr. W. Nocke for advice on the technique of the preparation and hydrolysis of 17-*epi*œstriol–acetonide and to Mrs. Brigitta Dumont and Miss Gerta Pangels for skilled technical assistance.

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[1] Marrian, G. F., Watson, E. J. D., and Panattoni, M., *Biochem. J.*, 65, 12 (1957).

[2] Brown, J. B., and Marrian, G. F., *J. Endocrinol.*, 15, 307 (1957).

[3] Breuer, H., Nocke, L., and Knuppen, R., *Z. physiol. Chem.*, 311, 275 (1958).

[4] Marrian, G. F., *Biochem. J.*, 24, 435 (1930).

[5] Marrian, G. F., and Bauld, W. S., *Biochem. J.*, 59, 136 (1955).

[6] Breuer, H., and Pangels, G., *Biochim. Biophys. Acta*, 36, 572 (1959).

Labelling of Deoxyribose with Carbon-14 in Relation to Incorporation of Phosphorus-32 into the Nucleotides of Deoxyribonucleic Acid of Rat Thymus Cells

It has previously been shown[1,2] that when rat thymus cells are incubated in the presence of glucose labelled with carbon-14, the isotope is incorporated to a different extent in the ribose moieties of the four individual types of nucleotide derived from ribonucleic acid. The ribose attached to the pyrimidine bases has a specific activity lower than that of the purine-bound ribose. The present report deals with experiments on the sugar component of deoxyribonucleic acid, and also with a comparison of carbon-14-labelling of deoxyribose and incorporation of phosphorus-32 into the nucleotides of deoxyribonucleic acid.

Male albino rats weighing about 150 gm. were used. The thymus glands were removed under ether anæsthesia; a suspension of the thymus cells was prepared[3] and the cells were incubated in the presence of glucose-^{14}C (generally labelled). At the end of the incubation the acid-soluble material and lipids were extracted from the thymus cells. The residue was extracted with 10 per cent sodium chloride solution at 100° C. to isolate the sodium nucleates, which were then fractionated into ribonucleotides and deoxyribonucleic acid by alkaline hydrolysis and subsequent acidification[4]. The deoxyribonucleic acid was degraded by deoxyribonuclease and snake venom to nucleosides which were separated by paper chromatography in *n*-butanol/ammonia[5]. Each of the nucleoside fractions was degraded[6] to give free deoxyribose, which was further purified by paper chromatography using ethyl acetate/acetic acid/water as solvent[7]. Deoxyribose was determined in an aliquot of the eluate by the diphenylamine reaction[8], and another aliquot was taken for radioactive assay.

Table 1 shows the specific activity of deoxyribose derived from the four nucleosides obtained from deoxyribonucleic acid. It can be seen that the incorporation of carbon-14 into the purine-bound deoxyribose is more than twenty times greater than the incorporation into the pyrimidine deoxyribose. This finding shows a similar trend to the results obtained with ribonucleic acid[2], although the difference between the values of specific activity of the purine ribose and the pyrimidine ribose was not as great as that found here for deoxyribose.

Table 1. Incorporation *in vitro* of Carbon-14 from Labelled Glucose into the Deoxyribose of Deoxyribonucleic Acid of Rat Thymus Cells

Source of deoxyribose	Specific activity of deoxyribose (counts/min./μmole)	Relative incorporation of carbon-14 into deoxyribose
Adenine deoxyriboside	325	100
Guanine deoxyriboside	420	129
Cytosine deoxyriboside	10	3
Thymidine	21	6

Each of two incubation vessels contained 6 ml. of cell suspension, obtained from eight thymus glands, in Krebs–Ringer phosphate solution (ref. 9) gassed with oxygen and containing glucose-^{14}C (generally labelled). Glucose concentration: 5 mgm./ml.; specific activity of glucose: $9{\cdot}89 \times 10^4$ counts/min./μmole. The vessels were shaken at 37·5° C. for 3 hr., the evolved carbon dioxide being absorbed in potassium hydroxide. At the end of incubation the contents of the two vessels were pooled.

In order to test whether the turnover of the phosphorus moiety of each of the deoxyribonucleotides follows a pattern similar to that of the corresponding deoxyribose moiety, experiments were carried out to determine the incorporation of phosphorus-32 into the four different types of nucleotide derived from deoxyribonucleic acid. Rat thymus cells were incubated in the presence of orthophosphate labelled with phosphorus-32. Deoxyribonucleic acid was isolated as described above and was degraded[10] by deoxyribonuclease and phosphodiesterase from rattle-snake venom[11]. The deoxyribonucleotides were separated using the paper electrophoresis technique of Markham and Smith[12], and their specific activities were then determined. The results of a typical experiment are shown in Table 2. It is clear that there is no difference between the incorporation of phosphorus-32 into the purine and the pyrimidine nucleotides, the specific activities of the four deoxyribonucleotides being of the same order. This finding is in sharp contrast to the observations made on the distribution of carbon-14 in the different deoxyribose moieties (Table 1), and also to the results reported for the sugar of ribonucleic acid where great differences were found between the specific activities of the ribose moieties of the different nucleotides[2].

The striking feature of these observations is the difference in pattern of incorporation of carbon-14 into the deoxyribose moieties of deoxyribonucleic acid and the incorporation of phosphorus-32 into the corresponding nucleotides. The difference in the degree of labelling of these two different moieties of a given nucleotide is of special interest in the study of the metabolism of deoxyribonucleic acid with the

Table 2. Incorporation *in vitro* of Phosphorus-32 into the Nucleotides derived from Deoxyribonucleic Acid of Rat Thymus Cells

Nucleotide isolated	Specific activity (counts/min./μmole)	Relative phosphorus-32 incorporation
Deoxyadenylic acid	465	100
Deoxyguanylic acid	576	124
Deoxycytidylic acid	502	108
Thymidylic acid	647	139

The incubation vessel contained 11 ml. of cell suspension, obtained from six thymus glands, in Krebs–Ringer bicarbonate gassed with 95 per cent oxygen – 5 per cent carbon dioxide (ref. 9). Orthophosphate labelled with phosphorus-32 was added to give a final concentration of 1 μc./ml. Glucose concentration was 5 mgm./ml. The vessel was shaken at 37·5° C. for 3 hr.

aid of labelled precursors. It indicates corresponding differences in the rates of turnover of the two moieties, the renewal of which presumably occurs while the nucleotides form part of the pool of free nucleotides of the cell constituting the precursors of deoxyribonucleic acid. It should be emphasized that the relative rates of turnover of these nucleotides, at least *in vitro*, evidently depend upon the moiety being studied.

It is not yet known whether similar differences in the pattern of incorporation of the two isotopes exist also between the nucleotides of ribonucleic acid; work is being carried out on this particular problem.

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[1] Itzhaki, S., paper presented at Fourth Int. Congr. Biochem., Vienna, Supp. to Int. Abst. Biol. Sci., Abstr. 6–55 (Pergamon Press, London, 1958).

[2] Itzhaki, S., *Biochim. Biophys. Acta*, **37**, 160 (1960).

[3] Kit, S., and Barron, E. S. G., *Endocrinol.*, **52**, 1 (1953).

[4] Davidson, J. N., and Smellie, R. M. S., *Biochem. J.*, **52**, 599 (1952).

[5] Tamm, C., Shapiro, H. S., Lipshitz, R., and Chargaff, E., *J. Biol. Chem.*, **203**, 673 (1953).

[6] Itzhaki, S. (to be published).

[7] Jermyn, M. A., and Isherwood, F. A., *Biochem. J.*, **44**, 402 (1949).

[8] Dische, Z., in "The Nucleic Acids", ed. by Chargaff, E., and Davidson, J. N., 1, 287 (Academic Press, New York, 1955).

[9] Umbreit, W. W., Burris, R. H., and Stauffer, J. F., "Manometric Techniques and Tissue Metabolism", second ed., 119 (Burgess Pub. Co., Minneapolis, 1949).

[10] Cohn, W. E., in "Methods in Enzymology", ed. by Colowick, S. P., and Kaplan, N. O., 3, 728 (Academic Press, New York, 1957).

[11] Koerner, J. F., and Sinsheimer, R. L., *J. Biol. Chem.*, **228**, 1049 (1957).

[12] Markham, R., and Smith, J. D., *Biochem. J.*, **52**, 552 (1952).

ANIMAL PHYSIOLOGY

Failure of Growth in Disturbed Laboratory Rats

Laboratory rats can be very sensitive to disturbances of their accustomed environment or routine, and they may react by modifying their eating and drinking behaviour in various ways[1-4]. If some kinds of disturbance are prolonged, the body-weight may be affected, and we have previously found this a particularly useful measure: rats which were exposed to disturbances either lost weight or they failed to gain it as quickly as undisturbed controls[5,6].

In one of these investigations[6] adult rats were subjected to disturbances of a kind which often occurs incidentally during laboratory experiments: transfer to an unfamiliar room and cages, separation from cage mates, a new form of diet, oral injections. These changes were introduced concurrently, and by the end of the next 24 hr. the rats had eaten and drunk only about half their normal intake and had lost nearly 3 per cent of their body-weight. The environmental changes were maintained for three weeks; by then the rats had not yet made good the loss of weight: a control group had meanwhile gained more than 3 per cent.

Relatively commonplace events seemed, therefore, to have evoked a big reaction, and in the present investigation this kind of situation was analysed further and over a longer period of time. In particular, the aim was to study effects on body-weight: (*a*) when environmental changes were introduced gradually, that is, one by one, and time was allowed for the rats to become adapted before the next change was made; (*b*) during a subsequent period when no further changes were made and the rats were left to 'recover' undisturbed.

Male Lister hooded rats lived under uniform conditions, three to a cage, since weaning and were little handled. When they were 200–210 days old they were allocated at random to two groups of five rats. The 'undisturbed' control group remained in their home cages in the colony room throughout and were merely weighed daily. The 'disturbed' experimental group were also weighed daily, and in addition they were treated as follows:

(1) *Transfer to another room.* The rats stayed in their home cages but were transferred by trolley to another room nearby where the cages were again suspended in standard racks. The temperature (set by thermostat at 68° F.) and the general level of light and noise in the new room were roughly the same as in the colony room; humidity could not be controlled but the fluctuations when measured were fairly similar in the two rooms. The usual diet (41.*b*, pellets) and water continued to be available *ad lib.*

(2) *Change of diet.* Eight days later, a powdered form of diet 41.*b* was substituted for the pellets.

(3) *Segregation.* After a further 8 days, the rats were separated from their cage mates and were re-housed singly in unfamiliar cages of identical pattern.

(4) *Injections.* After 8 days of living singly, and daily thereafter, each rat was given orally by syringe 1 ml./kgm. tapwater.

(5) *Daily change of cage.* Eight days after the injections had begun, each rat was re-housed in an unfamiliar though otherwise similar cage each day. Thus, once an environmental change had been introduced it was maintained, so that by now the rats were living in the new room, on powder food, singly, with a daily injection and in a different cage each day.

(6) '*Recovery.*' After 12 days, no further changes were introduced, injections ceased, and the rats remained—still singly and with powder food—in the same cage for a final 16 days. Throughout the investigation the body-weights and food and water intake were determined, and any environmental changes were made daily between 2.30 and 3.30 p.m. The mean starting weights were: experimental group 317·4 gm., *S.D.* 33·6, controls 323·2 gm., *S.D.* 45·8, and the difference was not significant ($P > 0·05$). All rats seemed in normal physical health throughout.

Fig. 1 shows the mean gains or losses in body-weight for the two groups during the 60 days. The 'undisturbed' controls gained weight fairly steadily throughout, and by the end the average gain was 29·0 gm., *S.D.* 5·4, or about 9 per cent. In the experimental group, however, weights changed relatively little during the first 44 days while the environmental disturbances were taking place. Immediately after all but one of the disturbances there was if anything a slight drop in weight, of the order of 2–3 gm., but this had usually been made good by the time the next kind of disturbance was due; the greatest drop followed the introduction of the new diet, but even this was not statistically significant. The pattern of daily food and water intake roughly reflected the fluctuations in weight; the greatest drop occurred, again, just after the change of diet when the average

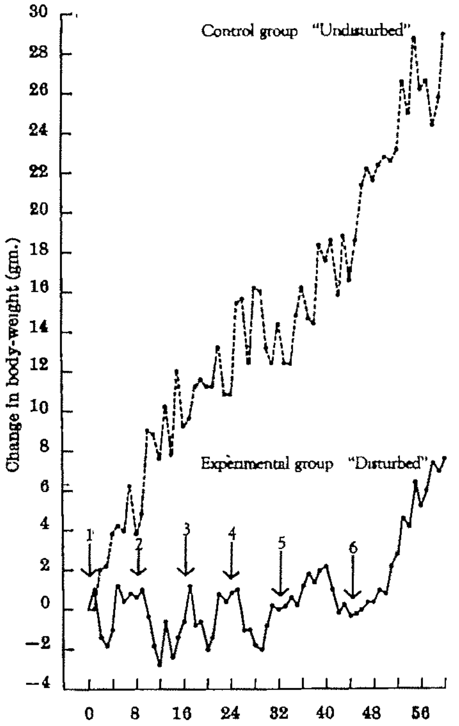

Fig. 1. Arrest of gain in weight in rats subjected to environmental changes. The control group remained undisturbed in the colony room throughout. The experimental group had successively the following treatments: (1) transfer to another room; (2) change in the form of diet; (3) segregation from cage mates; (4) daily oral injections; (5) daily change of cage; (6) 'recovery', that is, no further alterations in the environment were made. The arrows indicate when each kind of treatment began. Weight changes are expressed as mean gains or losses from the mean body-weight of each group at the beginning of the experiment

food and water intake was about a quarter less than usual. Thus at the end of the 'disturbed' period the experimental group still weighed almost exactly the same as at the beginning (mean change − 0·4 gm., *S.D.* 4·2); the controls had meanwhile gained 16·6 gm., *S.D.* 2·4. The net difference between the two groups was therefore 17·0 gm., and these changes in mean weight differ significantly between the two groups ($t = 7{\cdot}05$, $P < 0{\cdot}001$). When 'recovery' began, the weight of the experimental group immediately began to climb, and the graph suggests that they gained weight at a rate roughly parallel to that of the controls. By the end of the 16 days recovery they had gained 8·0 gm., *S.D.* 1·1, slightly less than the corresponding 12·4 gm., *S.D.* 3·9, gained by the controls. The net difference between the two groups at the end of the experiment was therefore 21·4 gm. ($t = 6{\cdot}14$, $P < 0{\cdot}001$), and this makes it seem unlikely that the experimental group was catching up. Food and water intake during the recovery period were very slightly higher than before but did not increase as clearly as body-weight.

These results suggest that the environmental changes led to a temporary arrest in the rats' normal weight gain. Since for practical reasons the order in which the changes were made was the same for all the rats, it is not strictly valid to compare the effects of the different kinds of change with each other. Superficially the effects of each kind of change were fairly similar, and the effect of the change of diet was possibly the greatest. Merely to move the rats from the colony room to another room at the beginning of the experiment (Fig. 1 (1)) had some effect, though this was smaller than the effect previously referred to of making various other changes concurrently[5].

A probable reason for the arrested gain in weight was that the disturbed rats temporarily ate and drank less than usual. Environmental changes of the kind described might, however, also have had other effects, like increased general activity[7,8], less-efficient assimilation of food[9], or loss of heat because of single living[10], and any of these could have helped to keep down body-weight. The experimental group was also, inevitably, handled slightly more than the controls, but handling probably benefits body-weight[11]. Whatever the exact explanation, the results illustrate that common laboratory procedures to which normally little importance might be attached can—at least where rats' body-weight is concerned—have sizeable and lasting adverse effects.

Effects of this kind can sometimes be mitigated by administering small doses of the tranquillizing drug chlorpromazine[6].

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[1] Miller, G. A., and Viek, P., *J. Comp. Psychol.*, 37, 221 (1944).
[2] Hebb, D. O., "The Organization of Behaviour", 193 (Wiley, New York, 1949).
[3] Chance, M. R. A., and Mead, A. P., *Behaviour*, 8, 174 (1955).
[4] Weiss, K., and Hurwitz, H. M. B., *Nature*, 183, 344 (1959).
[5] Steinberg, Hannah, and Watson, R. H. J., *J. Physiol.*, 147, 20*P* (1959).
[6] Steinberg, Hannah, and Watson, R. H. J., in "Neuro-Psychopharmacology". ed. Bradley, P. B., Deniker, P., Radouco-Thomas, C., 415 (Elsevier, Amsterdam, 1959).
[7] Lane-Petter, W., *Brit. J. Animal Behav.*, 1, 124 (1953).
[8] Berlyne, D. E., *J. Comp. Physiol. Psychol.*, 48, 238 (1955).
[9] Ruegamer, W. R., Bernstein, L., and Benjamin, J. D., *Science*, 120, 184 (1954).
[10] King, J. A., and Connon, Helen, *Physiol. Zool.*, 28, 233 (1955).
[11] Bovard, E. W., *Psychol. Rev.*, 65, 257 (1958).

Modification of Cholesterosis and Lipidosis of Rats maintained on an Atherogenic Diet

Although the liver is regarded as the primary organ of lipid metabolism, little is known of this metabolic process at the cellular level. Recognizing that the cellular composition of liver is approximately 61 per cent parenchymal and 33 per cent littoral cells[1], I attempted to clarify the participation of hepatic cells in lipid metabolism by studies on the lipid composition of isolated parenchymal and Kupffer cells[2]. This work showed, in reference to the isolated parenchymal cell, an elevated cholesterol-level in Kupffer cells which suggested that these cells actively participated in cholesterol metabolism. Subsequent work indicated that Kupffer cells probably are not involved in the uptake of plasma cholesterol but do participate in its biotransformation or excretion[3].

The present work was designed to investigate the relationship between reticuloendothelial activity, and the ability of the animal to handle an exogenous cholesterol load. If Kupffer cells are involved in the biotransformation or excretion of cholesterol, then during reticuloendothelial hyperactivity one would anticipate increased tolerance to ingested cholesterol.

Table 1. INFLUENCE OF ZYMOSAN ON HEPATIC LIPIDS* OF NORMAL AND CHOLESTEROL-FED RATS

Group	No.	Body-weight (gm.)		Liver (per cent body-weight)	Spleen (per cent body-weight)	Cholesterol			Neutral fat
		Initial	Final			Free	Ester	Total	
Control	9	209 ± 6 6	265 ± 11·8	3·7 ± 0·19	0 23 ± 0·01	1 82 ± 0 08	0·71 ± 0·11	2 53 ± 0·10	9 14 ± 2·25
Control and zymosan	9	206 ± 11 5	211 ± 6·1	6 3 ± 0·24	1·10 ± 0 18	2·18 ± 0·09	0·35 ± 0 12	2 54 ± 0·14	6·81 ± 0·71
Cholesterol-fed	9	201 ± 6·6	198 ± 13 2	5 4 ± 0 25	0·24 ± 0·01	4·60 ± 0·29	50·56 ± 4 25	54·57 ± 4·55	38 93 ± 3·34
Cholesterol-fed and zymosan	9	212 ± 6·8	168 ± 4·9	7·1 ± 0 48	0·78 ± 0 09	3·45 ± 0·15	17·20 ± 2·74	20·98 ± 2·77	17·89 ± 1·82

* Lipid values are expressed as mgm./gm. of fresh tissue weight ± standard error.

This would be reflected by lower tissue cholesterol-levels. Since reticuloendothelial blockage cannot be produced[4], the influence of reticuloendothelial hyperactivity on liver cholesterol-levels was studied in rats maintained on normal and high cholesterol, high fat diets. The reticuloendothelium was stimulated by the intravenous injection of the yeast extract, zymosan, in the amount of 10 mgm. administered on alternate days. Thirty-six male rats were initially employed, 9 normal rats served as controls and received saline injections, while 9 additional normal rats received zymosan. The remaining 18 rats, 9 of which received zymosan, were fed an atherogenic diet similar to that employed by O'Neal *et al.*[5], which contained the following :

	Per cent		Per cent
Casein 'vitamin free'	19·9	Thiouracil	0 3
Sucrose	18·1	Choline chloride	0·2
Salt mix (U.S.P. XIV)	7 3	Sodium cholate	1·8
Alphacel (non-nutritive bulk)	9·1	Butter	37 0
Vitamin diet fortification mixture	1·8	Cholesterol	4·5

Daily determinations were made on body-weight and food-intake. At the end of the 14-day period liver total and free cholesterol, phospholipid and neutral fat were determined as previously described[6].

In agreement with previous observations[7] the administration of zymosan to rats maintained on 'Purina' show significantly increased weight of liver and spleen. The liver free and ester cholesterol fractions were not significantly altered in the normal rats given zymosan. Maintenance of rats on the atherogenic diet for 14 days significantly elevated the free cholesterol and profoundly increased the ester cholesterol and neutral fat. The administration of zymosan to rats maintained on the diet induced a comparable increase in liver and spleen weight despite the elevated dietary thiouracil level. Zymosan significantly reduced the accumulation of free and ester cholesterol and neutral fat in the liver. No correlation could be demonstrated between changes in body-weight and liver cholesterol-levels. Dietary-intake was not significantly different in the saline- and zymosan-treated groups maintained on the atherogenic diet. A significant reduction in plasma ester cholesterol was also noted in the zymosan-treated group, although no change was observed in the concentration of various lipid fractions of the spleen.

A pair-feeding experiment was also carried out for a 5-day period with 6 male rats maintained on 'Purina' chow to which 1 per cent cholesterol and 0·5 per cent sodium cholate was added. This would eliminate any question of body-weight alterations and also evaluate the effectiveness of zymosan in preventing liver cholesterol accumulation at lower levels of dietary cholesterol intake. These animals lost a mean 9 per cent of their initial weight. In the untreated cholesterol-fed group the liver ester cholesterol concentration was significantly elevated (Table 2). As observed in the previous studies, a significant reduction (88 per cent) occurred in the ester cholesterol fraction of the rats which were daily injected intravenously with zymosan in the amount of 5 mgm./day.

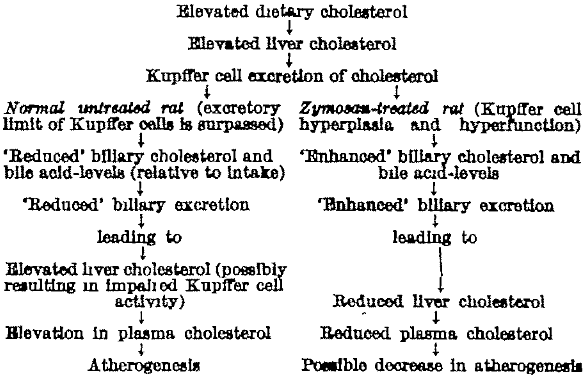
It is apparent that a major determinant to elevated plasma and liver cholesterol-levels of rats maintained on a high cholesterol diet is the functional state of the reticuloendothelium and specifically the number and activity of Kupffer cells.

Table 2. EFFECT OF ZYMOSAN ON LIVER CHOLESTEROL-LEVELS* OF PAIR-FED RATS ON A 1 PER CENT CHOLESTEROL DIET

Group	Body-weight (per cent)		Cholesterol		
	Liver	Spleen	Total	Free	Ester
Normal	3·41	0 19	2·16	1·91	0 25
Cholesterol-fed	3 01	0 24	18·03	3·20	14 83
Cholesterol-fed + zymosan	3 95	0 69	4 48	2 26	1·82

* Mean values are expressed as mgm. of cholesterol per gm. of fresh tissue.

Stambul[8] has previously summarized evidence for the role of Kupffer cells in the biliary excretion of cholesterol. In view of our previous observations[2,3] and the known mode of excretion of cholesterol which in the rat is an array of bile acids, the following concept can be proposed. This concept indicates the possible mechanism whereby zymosan prevents the accumulation of liver and plasma cholesterol and the possible etiology of hypercholesterolæmia and lipidosis in the untreated animals :


Elevated dietary cholesterol
Elevated liver cholesterol
Kupffer cell excretion of cholesterol
Normal untreated rat (excretory limit of Kupffer cells is surpassed)
Zymosan-treated rat (Kupffer cell hyperplasia and hyperfunction)
'Reduced' biliary cholesterol and bile acid-levels (relative to intake)
'Enhanced' biliary cholesterol and bile acid-levels
'Reduced' biliary excretion
'Enhanced' biliary excretion
leading to
leading to
Elevated liver cholesterol (possibly resulting in impaired Kupffer cell activity)
Reduced liver cholesterol
Elevation in plasma cholesterol
Reduced plasma cholesterol
Atherogenesis
Possible decrease in atherogenesis

In view of the functional complexity of the reticuloendothelium, and the role of zymosan in immunological mechanisms, other phases of reticuloendothelial activity besides reticuloendothelial involvement in cholesterol metabolism could be the mechanism of the protective action of zymosan. However, the present findings apparently demonstrate a new concept in the regulation and prevention of hepatic lipidosis and cholesterosis and possibly experimental atherosclerosis through the activation of the reticuloendothelium.

Work to substantiate further this hypothesis is now in progress.

The accumulation of lipid in Kupffer cells is reported to be most marked in diabetes[9], a condition associated with impaired reticuloendothelial activity[10] as well as hyperlipæmia and increased incidence of atherosclerosis.

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[1] Daoust, R., "Liver Function", pp. 3–11, ed. by Brauer, R. W. (Amer. Inst. Biol. Sci., Washington, D.C., 1958).

[2] Di Luzio, N. R., *Amer. J. Physiol.*, 196, 884 (1959).

[3] Di Luzio, N. R., *J. Amer. Oil Chemists* (in the press).

[4] Di Luzio, N. R., Simon, K. A., and Upton, A. C., *Arch. Path.*, 64, 649 (1957).

[5] O'Neal, R. M., Thomas, W. A., and Hartroft, W. S., *Amer. J. Cardiol.*, 3, 94 (1959).

[6] Powers, B. S., and Di Luzio, N. R., *Amer. J. Physiol.*, 195, 166 (1958).

[7] Gorstein, F., and Benacerraf, B., *Fed. Proc.*, 18, 478 (1959).

[8] Stambul, J., "The Mechanisms of Disease" (Froben Press, Inc., New York, 1952).

[9] Roessle, R., *Verhl. Deutsch. Path. Ges.*, 10, 334 (1907).

[10] Juhlin, L., *Acta Physiol. Scand.*, 45, 369 (1959).

Diathelic Immunization of Sheep against a Rat Cancer

THE term 'diathelic immunization' has been introduced by B. Campbell *et al.*[1], who instilled different antigens into the orifice of the milk gland (in cows) and obtained colostrum and milk with high titres of antibodies against the antigens used. They consider the milk gland as an exocrine reticulo-endothelial gland.

It has been shown by previous work that resistant rats (strain *bWr*, or black-Walker-resistant, inbred in our laboratory) produce, after implantation of the Walker 256 tumour, factors inhibiting the growth of this tumour in susceptible rats, when resistant immune blood serum is injected into them intraperitoneally[2], and that susceptible rat sucklings are protected against the growth of the tumour by the ingested milk when nursed by the resistant immune rat females[3].

Further, it has been shown that spleens of resistant rats immunized by the tumour *in vivo* continue the production of antitumour factors when explanted in tissue culture[4], and that an active immunization *in vitro* of spleens from normal resistant rats against the tumour used is possible[5].

In the present experiments, we instilled repeatedly, at weekly intervals, by means of a glass canule, several million of live Walker 256 tumour cells suspended in about 0·5 ml. of saline into the orifice of the milk glands of two sheep a month or more after parturition. Instillation was carried out on the evening after the last (third) day's milking. 12 hr. later (on the next morning) the milking was continued.

There was a massive cellular reaction in the milk from the instilled udders, changing gradually, in the course of the immunization, from nearly exclusively polymorphonuclear (segmented) leucocytes to more and more mononuclear cells with a large plasmatic component. (This cellular reaction will be described in detail and analysed in a separate communication.) There was also a strong decline in the milk output beginning at 18 hr. after instillation, but in the following 3–5 days milk output returned nearly to normal. There were no signs of bacterial inflammation of the instilled udders or of the animals themselves, although some capsulated bacteria have been seen in cell-preparations from the milk; milking was done under the usual non-aseptic conditions. The cellular reaction as well as the inhibition of lactation seem to vary from individual to individual; both were much stronger in one sheep than in the second.

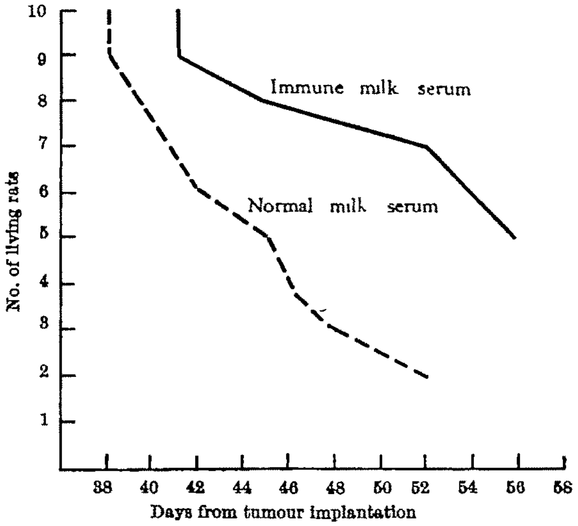

Fig. 1. Deaths from tumour in rats injected with normal and immune milk serum

The 'immune' milk was used in several ways.

(1) A neutralization test was performed. Rats were implanted with equal doses of the Walker 256 tumour cells, which had been incubated for 3 hr. at 37° C. in the normal and immune milk (only the milk fat was separated by centrifugation). After incubation, the cells were centrifuged from the milk and resuspended in saline before implantation. A control set of rats was implanted with non-incubated tumour cells (from the same original suspension), in saline. Results are given in Table 1.

Table 1

Incubation	Tumours*	
None	7/10	(70 per cent)
In normal milk	17/20	(85 per cent)
In immune milk	9/20	(45 per cent)

* Ratios of rats with tumours to the total number of animals used; after 30 days no new tumours appeared.

(2) Milk serum was prepared by adding some drops of dairy rennet and centrifuging the curdled milk, and injected after Seitz-filtration intraperitoneally into susceptible rats bearing implants of equal numbers of live Walker 256 cells. Results of two groups of 10 male rats weighing 120–150 gm. at the start of the experiment, and given, from the day of tumour implantation, 2 ml. daily of normal or immune milk serum for 40 days, are illustrated in Fig. 1. In the normal series there are 2 non-takes in 10, in the immune series 5 non-takes in 10.

It is to be noted that the gains in weight of rats given the immune milk serum were lower than of those given the normal milk serum or of the controls. The gains in weight of different groups after 15 days from the beginning of the experiment are given in Table 2.

Table 2

Injections	Mean gain in weight
None	16 per cent
Normal milk serum	15 per cent
Immune milk serum	8 per cent

After 15 days, the weight control is obscured by the growing tumours. On the whole, the anti-rat component of the heterologous (sheep) immune milk under given conditions seems negligible; all rats surviving without tumours, both in the experimental as well as in the control groups, seem undamaged and live normally to date.

(3) Cytotoxic effect of the immune milk serum was proved by the method of unstained cell count[6]. Freshly prepared tumour cells were suspended in normal and immune milk serum, counted, incubated at 37° C. for 4 hr. and counted again. Table 3 indicates results of these counts on 6 subsequent days, where the influence of milk sera from three different sheep after different numbers of immunizations are compared. In all these experiments, drops of complement were added (guinea pig serum); but we have found that fresh immune milk serum has a cytotoxic activity of equal degree without the addition of complement.

Table 3

Sheep No.	No. of immunizations	Cell count (per cent) before incubation	Cell count (per cent) after incubation, mean from 6 days	Range
1	none	100	96	82–100
2	3	100	57	48–67
3	5	100	18	10–26

These results, considered with previous work, seem to prove that any system capable of immune reaction is able to produce anti-tumour factors passively transmissible, and active in inhibiting tumour growth in susceptible animals. Furthermore, there seems to be a possibility of producing relatively large amounts of these active substances by a procedure which is very simple and obviously harmless for the producing animal. Thus some kind of immunotherapy could be used in combination with other therapeutic means in cancer.

Attempts to separate the active substances from the milk serum (as milk globulins), as well as to purify them from the anti-species component, are proceeding. Also the possible role of the cellular milk reaction and the eventual use of such cells are being considered.

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[2] Sekla, B., and Barvič, M., *Nature*, 178, 497 (1956).

[3] Sekla, B., *Nature*, 181, 1600 (1958).

[4] Holečková, E., and Sekla, B., *Nature*, 180, 1298 (1957); *Neoplasma* (Bratislava, ČSR), 5, 20 (1958).

[5] Sekla, B., and Holečková, E., Proc. Eighth Int. Cancer Congr., London, 1958, in *Acta Union Intern. contre le Cancer*, 15, 976 (1958).

[6] Schrek, R., *Amer. J. Cancer*, 28, 889 (1936).

Fibrinolytic Activity of Cirrhotic Liver

Several investigators have observed increased fibrinolytic activity in the circulating blood in cases of liver cirrhosis in man[1–4]. The cause of this activity is unknown, though a toxic effect similar to that produced in dogs by chloroform or peptone has been suggested. Such a mechanism could possibly be involved in an acute process, but could barely explain the prolonged periods of increased fibrinolytic activity observed in cases of liver cirrhosis.

A mechanism by which the fibrinolytic system in blood is activated by an activator of plasminogen released from injured tissue is apparently excluded, since such fibrinolytic activity has been reported to be absent from liver tissue[5–8].

We have now investigated the fibrinolytic activity of tissue from a series of cirrhotic livers, estimating the concentrations of plasminogen activator by a quantitative method[9]. The results are given in Table 1. It is seen that the samples from the cirrhotic livers show varying degrees of fibrinolytic activity. When compared with the concentrations in normal human organs[8] some of the pathological liver samples even range among the most active tissues in the human organism. Another significant observation is a slight increase in activity of apparently normal livers with age (Table 1).

Table 1. Fibrinolytic Activity of Apparently Normal and Cirrhotic Human Livers estimated as Concentration of Plasminogen Activator in Units of a Standard Preparation per gm. Fresh Tissue

Normal group Age	Normal group Sex	Normal group Activity	Cirrhotic group Age	Cirrhotic group Sex	Cirrhotic group Activity
42	M	0	40	M	1,935
55	M	0	67	F	80
59	F	0	68	M	650
60	M	27	68	M	262
62	F	0	70	F	18
64	M	11	74	M	63
65	M	0	76	F	456
66	M	19	77	M	935
66	M	1	78	M	1,781
69	F	1	78	F	205
72	M	0	80	M	188
73	F	41			
74	M	1			
74	M	42			
74	M	0			
78	F	28			
80	M	75			
80	F	48			
82	F	160			
88	M	0			

In accordance with views previously presented[10] fibrinolysis participates in wound healing. There are now several indications that vascularized connective tissue, occurring normally in the organism or formed as response to tissue injury, is characterized by high fibrinolytic activity caused by a plasminogen activator. Thus normal animal and human meninges are among the most active tissues so far investigated[11]. High activity was also recorded in the adventitia of arteries[12] or in the walls of veins[13] as well as in the kidney capsula[14]. The activity of the arterial adventitia increases with age[15], probably reflecting increased vascularization. Similarly, high fibrinolytic activity was found in connective tissue from samples of healed human myocardial infarcts[16] in contrast to the fairly low activity of normal human myocardium. The last two examples apparently represent a response to tissue injury similar to that occurring in liver cirrhosis or produced during ageing. The lack of fibrinolytic activity of blood in cases of primary carcinoma of the liver[3] probably points to a disturbed tissue repair process.

Since plasminogen is probably formed in the liver, it is easy to visualize that dangerous conditions could arise when high concentrations of a plasminogen activator are found in close contact with plasminogen-producing liver cells. Apart from interfering with local wound healing and tissue repair, fibrinolytic activity could easily escape into the blood system. Hence, it is possible to understand the physiological basis of the morbidity of diseases leading to cirrhosis of the liver.

The results are also of interest in connexion with the low incidence of arterial disease recorded in patients with liver cirrhosis by some authors. The observations support the thrombogenic theory of the pathogenesis of arteriosclerosis, as set forth by Duguid[17,18], and strengthen the concept of a dynamic balance between fibrin formation and fibrinolysis as a regulator of this process[10,19].

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[1] Goodpasture, E. W., *Bull. Johns Hopkins Hosp.*, 25, 330 (1914).
[2] Ratnoff, O. D., *Bull. Johns Hopkins Hosp.*, 84, 29 (1949).
[3] Kwaan, H. C., McFadzean, A. J. S., and Cook, J., *Lancet*, i, 132 (1956).
[4] de Nicola, P., and Soardi, F., *Thromb. Diath. Haemorrh.*, 2, 290 (1958).
[5] Fleisher, M. S., and Loeb, L., *J. Biol. Chem.*, 21, 477 (1915).
[6] Permin, P. M., *Acta Physiol. Scand.*, 21, 159 (1950).
[7] Lewis, J. H., and Ferguson, J. H., *J. Clin. Invest.*, 29, 1059 (1950).
[8] Albrechtsen, O. K., *Brit. J. Haematol.*, 3, 284 (1957).
[9] Astrup, T., and Albrechtsen, O. K., *Scand. J. Clin. Lab. Invest.*, 9, 233 (1957).
[10] Astrup, T., *Lancet*, ii, 565 (1956).
[11] Moltke, P., *Proc. Soc. Exp. Biol. Med.*, 98, 377 (1958).
[12] Astrup, T., and Claassen, M., Trans. European Soc. Hæmatol. Sixth Congr., Copenhagen 1957. 455 (Karger, Basle).
[13] Todd, A. S., *Nature*, 181, 495 (1958).
[14] Ladehoff, Å., *Scan. J. Clin. Lab. Invest.* (in the press).
[15] Astrup, T., Albrechtsen, O. K., Claassen, M., and Rasmussen, J., *Circulation Res.*, 7, 969 (1959).
[16] Fischer, S., Albrechtsen, O. K., and Bang, N., *Thromb. Diath. Haemorrh.*, 3, 554 (1959).
[17] Duguid, J. B., *J. Path. Bact.*, 58, 207 (1946); *Brit. Med. Bull.*, 2, 36 (1955).
[18] Morgan, A. D., "The Pathogenesis of Coronary Occlusion" (Blackwell, Oxford, 1956).
[19] Astrup, T., in Page, Irvine H., "Connective Tissue, Thrombosis, and Atherosclerosis", 223 (Academic Press, New York, 1959).

Rapid Contractile Properties of Isolated Mammalian Arteries

STRONG indirect evidence is now accumulating[1,2] in support of the old idea that large mammalian arteries may be capable of contracting and relaxing at least as rapidly as the heart. Direct evidence based on studies of isolated arteries is still lacking, and this remains a major stumbling block in accepting the implications of much of the recent work.

Paton's technique[3] has been used to study the behaviour of 2–3 cm. lengths of dog's common carotid, femoral, hepatic and splenic arteries, and of sheep's hepatic and splenic arteries. The excised artery was tied on a glass cannula which formed a closed system connected to a manometer (Hansen condenser). The free end and any branches were ligated with fine silk, and the artery was stretched to about three-quarters of its natural length by a stiff silver wire passed through the cannula and along the lumen of the artery. This wire electrode was led through a seal into the air, and another silver electrode dipped into the bath in which the artery was immersed. Tyrode's solution at 37° C. was used in most experiments, and saturated with 5 per cent carbon dioxide and 95 per cent oxygen. From ½ to 4 hr. after excision all the 34 arteries examined were capable of raising the pressure within the closed system when an electrical stimulus was applied across the two electrodes. To square-wave stimuli the chronaxie was about 20 m.sec. and the rheobase 1–4 V. Responses were obtained equally easily whether the central lumen electrode was positive or negative. The highest rise of pressure obtained with a single shock (of an intensity and duration just less than that which caused irreversible damage to the preparation) was 15 mm. mercury. Much higher pressures could be obtained by repetitive stimulation. It was difficult to elicit any response if the starting pressure in the system exceeded 200 mm. mercury, and low pressures were used in most experiments because of the difficulty of stopping very small leaks.

The time course of a typical single contraction is shown in Fig. 1*A*, and the latency of onset of the contraction is shown at a faster film speed in Fig. 1*B*. Latency is 80 m.sec., time to peak pressure 2–3 sec., and time to full relaxation 5–10 sec. The speed of relaxation was usually greater at high internal pressures, at least within the physiological range. The contraction was reversibly abolished by increasing external potassium concentration to 130 m.equiv./l. (isosmotic with normal Tyrode solution); but it could still be elicited in the presence of atropine (10^{-7}) and dibenyline (10^{-6}) which had been allowed to act for 30 min. Slow repetitive stimulation produced pressure oscillations at frequencies up to about 0·7 c./s. (Fig. 2*A*). If an oscillating pressure was introduced into the system, the amplitude of oscillation was increased by stimulation (Fig. 2*B*), presumably because of increased rigidity of the muscular wall. The long chronaxie and the failure of drugs to influence contraction indicate that muscle cells are being excited directly. So far it has not proved possible to excite contraction through nerves, presumably because in this preparation the nerve supply is damaged. Even when arteries were tested in fresh plasma 5 min. after excision, there was no evidence at all of excitation of nerves. By analogy with other excitable tissues, it seems probable that

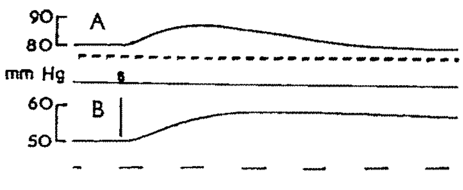

Fig. 1. Time-course of the contraction to a single stimulus (*S*) of a 2-cm. length of sheep's hepatic artery. (*A*) From above down: the stimulus (18 V., 20 m.sec., isolated from earth and fed to the cathode-ray oscillograph through capacitors); lumen pressure (scale on left in mm. mercury); time marking (½ sec.). (*B*) Onset of a contraction (at a faster film-speed) following a stimulus of 120 V. for 150 μsec.: time marking, ½ sec.

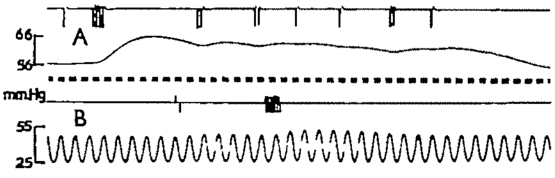

Fig. 2. (*A*) Response of a dog's hepatic artery to short bursts of repetitive stimulation. Top trace shows the stimulus record (retouched) and lower trace the pressure record. (*B*) Effect of an oscillating pressure introduced in the same preparation. Stimuli (upper trace, retouched) cause a rise of mean pressure and of oscillation amplitude (lower trace). Time marking (¼ sec.) in between the records is the same for both

excitation through the normal nerve supply will produce more rapid effects.

Since efferent sympathetic activity normally occurs in bursts with a cardiac rhythm[4], it seems possible that rapid response of large arteries to such nervous activity could explain a number of curious phenomena observed in the peripheral arterial pulse contour, notably the augmentation of systolic peak pressure towards the periphery and perhaps also the dicrotic wave. It is noteworthy that atropine and procaine can abolish both these phenomena[5], even though there may be no change in mean arterial pressure.

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[1] Wehn, P. S., *Acta Physiol. Scand.*, **46**, 107 (1959).
[2] Heyman, F., *Acta Med. Scand.*, **164**, Supp. 344 (1959).
[3] Paton, W. D. M., *J Physiol.*, **127**, 40,*P* (1955).
[4] Adrian, E. D, Bronk, D. W., and Phillips, G., *J. Physiol* **74**, 115 (1932).
[5] Jungmann, H., and Rohr, H., *Pflug. Arch. ges. Physiol.*, **258**, 38 (1953).

Thyroidal Control of Radiophosphorus Metabolism in Salmon

The part played by the thyroid gland in teleost metabolism has been investigated by many workers. In cold-blooded vertebrates it appears to be concerned more with growth than with general metabolism[1]. However, there is a great deal of controversy on practically every aspect of thyroid physiology. In higher vertebrates, the parathyroid plays an important part in the mineral metabolism. Fishes have, so far as known, no parathyroid. It has been suggested[2] that the ultimobranchial body in the teleost is the homologue of the parathyroid but as yet nothing positive has been established regarding this hypothesis. Fishes are able to control mineral metabolism within narrow limits; but there is scarcely any information concerning the role of the endocrines in this regulation. Radioisotopes are being used to carry out experiments to find out whether the thyroid gland has any influence on mineral metabolism in fishes.

In fishes the thyroid gland is divided into innumerable follicles in the region of the ventral aorta, hence surgical thyroidectomy is impossible. Yearling salmon, *Salmo salar* L., were divided into two groups of which one was maintained in 0·3 per cent solution of thiourea, referred below as treated. The other was used as control. The fishes were kept in thiourea solution for two weeks, the solution being changed every three days. After two weeks the fish were transferred to fresh aquaria to which sufficient phosphorus-32 had been added to give an activity of 2,500 counts per min. per ml. Aquaria for treated fish also contained 0·3 per cent solution of thiourea. At regular intervals starting from 2 hr. the tissues (liver, kidney, muscle and bone) were prepared for counting. The readings were corrected for decay and checked against a uranium standard. Carrier-free phosphorus-32 used was received from Atomic Energy of Canada, Ltd., in the form of phosphoric acid in diluted hydrochloric acid. In both the sets of experiments, bone took up the maximum activity, followed by liver, kidney and muscles. The uptake was always more in the treated set than the control. In the control, 40·1 per cent of phosphorus-32 was absorbed, whereas in the treated 50·3 per cent was utilized.

Another set of experiments was arranged to throw further light on the problem. The fishes were divided into three groups. One of them was injected with 0·2 ml. of 0·1 per cent solution of thiourea, the other with 0·2 ml. of 0·1 per cent solution of l-thyroxine and the third with 0·2 ml. physiological saline to serve as control. The injections were repeated again after three days. After a lapse of three days the fish were transferred to solutions of phosphorus-32 which gave an activity of 2,500 counts per min. per ml. In these experiments the uptake of radiophosphorus in bone and liver was studied. In the control, the fishes took up 39 per cent, fishes injected with thiourea absorbed 52 per cent and thyroxine-injected fishes utilized 63 per cent of phosphorus-32.

Thiourea had been used in the above experiments to destroy the thyroid gland and so to check its activity; but the histological studies showed that it was not achieved. Thyroid follicles were still present lined with low epithelial cells and were full of colloid. Administration of antithyroid drugs either abolishes or retards the ability of thyroid gland to synthesize thyroxine[3]. As a consequence of the cessation of thyroxine synthesis, the level of circulating hormone falls and the pituitary immediately responds by secreting additional amounts of thyrotropin which stimulates the thyroid so that hypertrophy of the gland results. This then shows that destruction of thyroid can only be achieved after treating the fishes with antithyroid drugs for considerably longer times.

The results of the experiments can be interpreted that in the fishes treated with thiourea the thyroid was stimulated indirectly (by thyrotropin secreted by the pituitary gland) and so the uptake of radiophosphorus was more than the control. In fishes treated with thyroxine the stimulation was direct and so the uptake was maximum. Detailed results of the experiments will be published elsewhere.

Complete thyroidectomy can be achieved by repeated doses of radioiodine[4]. Further work to study the effect of hypophysectomy and destruction of thyroid by radioiodine upon mineral metabolism is under investigation.

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[1] Lynn, W. G., and Wachowski, H. E, *Quart. Rev. Biol.*, **26**, 123 (1951).
[2] Rasquin, P., and Rosenbloom, L, *Bull. Amer. Mus. Nat. Hist*, **104**, 359 (1954).
[3] Astwood, E. B., Brookhaven Symp. Biol., **7**, 61 (1955).
[4] La Roche, G., and Leblond, C. P., *Proc. Soc. Exp. Biol. Med*, **87**, 273 (1954).

Use of Chorionic Gonadotropin to prevent Early Embryonic Mortality in Temporarily Fasted Mice

WHEN studying the pathogenesis of the infertility in female mice caused by depriving them of all food for short periods during early gestation[1], it was found that the immediate cause was either failure of implantation or death of the embryo after implantation. In the latter case, it appeared from the histopathology that the primary lesion occurred in the deciduoma. This was followed by the death of the embryo, possibly due to interference with its nutrition.

The minimal starvation required was the 48-hr. period from the fourth to sixth day after œstrus or third to fifth day after the formation of the copulation plug. Such fasting rendered all matings infertile. However, when injections of 5 I.U. of chorionic gonadotropin were given once daily to each of ten mice during the period when they were being starved, the embryos of all mice survived until at least the third day after the end of the fast. When examined at this time none of the twelve control mice, fasted but not injected, was pregnant. It was assumed from the results of earlier work that embryos had developed but had died and been reabsorbed. When chorionic gonadotropin was given to 16 mice in a third group, over the period from the start of starvation until five days after its end, the embryos of ten appeared normal on the sixth day after the end of fasting. The embryos of three were starting to degenerate and three mice were not pregnant. Progesterone in daily doses of 0·5 mgm. had a similar protective effect.

This suggests that the pathogenesis of the embryonic mortality involves the anterior pituitary–ovarian endocrine system.

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[1] McClure, T. J., *Nature*, 181, 1132 (1958); *J. Physiol.*, 147, 221 (1959).

Oxygen Consumption by Tissues from Rats injected with L- or D-Thyroxine

It has been shown by a number of investigators that L-thyroxine is more active *in vivo* than is D-thyroxine[1]. In most *in vitro* systems, however, the D- and L-isomers are equally effective. The equal activity of D- and L-thyroxine in altering the morphology of mitochondria and in 'uncoupling' oxidative phosphorylation are examples of this[2,3]. Previous experiments[4] in this laboratory have compared the distribution and excretion of labelled D- and L-thyroxine labelled with iodine-131 following a single injection of a tracer quantity of each compound into the tail vein of the rat. Initially, D-thyroxine was concentrated to a greater extent in the liver and kidney than was L-thyroxine, but after 24 hr. the concentrations were approximately equal. However, the concentration of D-thyroxine in tissues such as muscle, brain and skin was much less than the concentration of L-thyroxine. This difference in distribution to the peripheral tissues was of such magnitude as to suggest a possible basis for the different activities of the two isomers *in vivo*. Others have shown[5,6] that the oxygen consumption of certain tissues (for example, liver, kidney and muscle) from animals injected with DL-thyroxine is significantly greater than normal, whereas that of other tissues (for example, brain) is not increased. The current investigation was undertaken to determine whether the differences demonstrated in the distribution of the two isomers could be correlated with a difference in their ability to stimulate the oxygen consumption of the several tissues.

Adult male Wistar rats, 4–6 weeks post-thyroidectomy, were used in this experiment. The treated animals were injected subcutaneously for 4 days with either D- or L-thyroxine (6 mgm./kgm.). (D-thyroxine was provided through the kindness of Dr. L. G. Ginger, of Travenol Laboratories, Inc., Morton Grove, Illinois. L-thyroxine was furnished through the kindness of Dr. A. E. Heming, of Smith, Kline and French Laboratories, Philadelphia, Pa.) The sodium salt of each isomer was dissolved in 0·02 *N* sodium hydroxide and 0·9 per cent sodium chloride. All animals were fasted 24 hr. before being anæsthetized with ether and exsanguinated. The various tissues were rapidly excised and placed in ice-cold Krebs–Ringer–phosphate solution. All tissues were prepared in duplicate. Liver, kidney and heart slices prepared by the Deutsch technique[7] were made smaller by cutting with scissors. Each diaphragm was cleared of fat and connective tissue, and its central portion was removed; the hemi-diaphragms then were placed in separate flasks. Specimens of brain were prepared by removing the larger tracts of white matter and forcing the remaining tissue through a syringe and No. 18 needle. After weighing, the tissues were placed in chilled Warburg flasks containing the incubation medium (Krebs–Ringer solution buffered with 0·01 *M* sodium phosphate with 100 mgm. per cent glucose). The flasks were gassed with 100 per cent oxygen for 5 min.; oxygen consumption was determined at 37° C.

It may be noted in Table 1 that with slices of liver from animals injected with either D- or L-thyroxine the oxygen consumption is almost identical; in both cases the Qo_2 is significantly greater than that of the control. The same result was also obtained with kidney slices. With heart and diaphragm, on the other hand, there is a significant difference between the animals injected with L-thyroxine and those injected with D-thyroxine; in both tissues the Qo_2

Table 1. OXYGEN CONSUMPTION BY VARIOUS TISSUES FROM RATS INJECTED WITH L- OR D-THYROXINE

	Control Qo_2 (10 rats)	L-Thyroxine Qo_2 (10 rats)	D-Thyroxine Qo_2 (10 rats)	P Control vs. L-thyroxine	P Control vs. D-thyroxine	P L-Thyroxine vs. D-thyroxine
Liver	1·40 ± 0·06*	1·94 ± 0·03	1·91 ± 0·07	< 0·001	< 0·001	> 0·1
Kidney	3·03 ± 0·13	3·79 ± 0·19	3·84 ± 0·11	< 0·01	< 0·001	> 0·01
Heart	1·21 ± 0·06	1·51 ± 0·08	1·30 ± 0·04	< 0·01	< 0·001	< 0·001
Diaphragm	1·11 ± 0·03	1·48 ± 0·04	1·29 ± 0·04	< 0·001	< 0·001	< 0·01
Brain	1·66 ± 0·04	1·51 ± 0·04	1·60 ± 0·04	= 0·01	> 0·1	> 0·1

* Standard error of the mean, Qo_2 = cu.mm. of oxygen per mgm. wet weight per hr.

was higher with tissues from animals injected with the L-isomer. The Qo_2 of brain was slightly lower than control values in both series of treated animals. The present results support the thesis that the difference in physiological activity between the two isomers of thyroxine is probably secondary to their different distributions *in vivo*.

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[2] Tapley, D. F., *J. Biol. Chem.*, **222**, 325 (1956).
[3] Tapley, D. F., and Cooper, C., *J. Biol. Chem.*, **222**, 341 (1956).
[4] Tapley, D. F., Davidoff, F. F., Hatfield, W. B., and Ross, J. E., *Amer. J. Physiol.* **197**, 1021 (1959).
[5] Gordon, E. S., and Heming, A. E., *Endocrinol.*, **34**, 353 (1944).
[6] Barker, S. B., and Klitgaard, H. M., *Amer. J. Physiol.*, **170**, 81 (1952).
[7] Umbreit, W. W., Burris, R. H., and Stauffer, J. F., "Manometric Techniques and Tissue Metabolism" (Burgess, Minneapolis, 1952).

Pitch Discrimination in Orthoptera (Insecta) demonstrated by Responses of Central Auditory Neurones

For about twenty years, since Pumphrey's review of hearing in insects[1], the commonly accepted view has been that the auditory organs of insects convey to the central nervous system considerable data about the pattern of amplitude of impinging sounds. There has been, on the other hand, no good evidence that the primary sensory nerves can or must convey cues as to the pitch of a sound stimulus and there has been no good evidence that the central nervous system responds differentially to one pitch rather than another, irrespective of intensity.

By recording from the pro-mesothoracic connectives of *Locusta migratoria* and *Schistocerca gregaria* (Acrididae) or from the cervical connectives of *Acheta domestica* (Gryllidae) with external electrodes, it is possible to identify a few ascending internuncial neurones which are excited at the sudden onset of a pure tone or short pulse of sound. The receptor is the tympanal organ, as may be shown by section of its nerve. The curve of threshold strength plotted against stimulus pitch for the single post-ganglionic neurone with the lowest threshold does not correspond with the analogous curve obtained by recording the summed action potentials of the sensory nerve, as initiated by the same stimuli. When recording from the sensory nerve the threshold was taken to be the stimulus strength required to produce a response of a fixed height on the oscilloscope screen, about one-twentieth of the maximal response.

The threshold strength of sound pulses required to elicit a response in a post-ganglionic neurone is raised by the simultaneous sounding of a continuous note. At the same time the pitch range of maximum sensitivity is moved as shown in Fig. 1. The direction of the shift of optimum pitch is away from the pitch of the continuous note. These results have been obtained with each of the above species of Orthoptera.

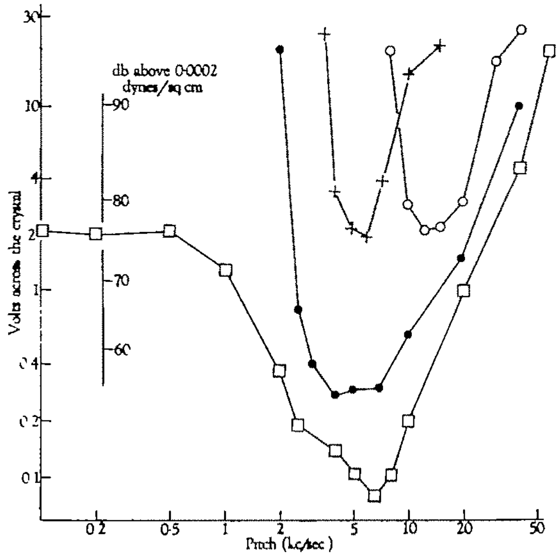

Fig. 1. Sound intensity threshold curves for auditory responses of *Locusta migratoria* with tone pulses of various pitch. □, From the tympanal nerve, the threshold being taken as the intensity required to give a twentieth of maximal response height. ●, From a single ascending neurone of the ventral cord. +, From the same neurone in the presence of a continuous tone at 15 kc./s. ○, From the same neurone with a continuous tone of 4 kc./s. Each point is the average of two readings, one taken ascending, the other descending the scale. The ordinate of sound intensity (db. above 0·0002 dynes/sq. cm.) is useful only up to 15 kc./s. and in the lower ranges is accurate to about 10 per cent

The difference between the pre- and post-ganglionic threshold curves is such that the metathoracic ganglion of the locusts responds differentially in favour of tone pulses of high pitch and rejects those of low pitch although they excite axons of the tympanic nerve. In the cricket, on the contrary, the prothoracic ganglion discriminates in favour of pulses in a low pitch-range of 0·5–3 kc./s., though the thresholds measured in the tympanic nerve do not begin to rise steeply until a range of 20–30 kc./s. is reached. Therefore, this evidence of discrimination indicates that the ganglion 'selects' certain axons, and this in turn means that discrimination has already occurred at the sense organ although formerly not observed by electrical recording from the whole bundle. The second piece of evidence, that the optimum pitch for the response to a pulse is shifted by a continuous note of differing pitch, does not indicate whether the mechanism lies in the ganglion or in the sense organ. A suggested explanation of these two sets of results is that there are at least two groups of sensory neurones, one group more easily excited by pulses of low pitch, one by pulses of high pitch. The group of neurones which become accommodated to a continuous note of one pitch would not then include those which are accommodated to a note of widely differing pitch.

Confirmation that different groups of sensory neurones are excited by the sudden onset of pure tone pulses of differing pitch comes from careful examination of the summed action potentials of whole tympanic nerves of the above two species of Acrididae. In preparations in which there is a favourable arrangement of the electrodes in relation to the short-circuiting of action currents, the summed action potentials initiated by pulses of one pitch cannot be matched in exact form by those initiated by pulses which differ only in pitch, no matter how relative stimulus intensities are adjusted. The response is of a number of axons in each of which there appears to be only one impulse when sound pulses of 10–20 m.sec. are used. Therefore sound pulses of differing pitch initiate impulses in distinct groups of sensory axons.

A possible mechanism for pitch discrimination is suggested by the structure of the tympanal sense organ of the above species. The sensory neurones are not situated all at the same point but are separated into groups. In agreement with the arguments of Pumphrey[1] the sense organ behaves as a displacement receptor without resonance, but it is still possible for pulses of differing pitch to initiate differing modes of vibration so that the groups of sensory neurones are not stimulated always in the same ratio. Such a mechanism would account for the small size of the observed effects. Certainly the mechanism of analysis is far inferior to that of the mammalian ear, but would provide improved discrimination of biologically important noises from a background disturbance.

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[1] Pumphrey, R. J., *Biol. Rev.*, 15, 107 (1940).

HÆMATOLOGY

Mechanism of Clot Lysis

The availability of a large number of inhibitors of proteolytic enzymes, as well as the availability of highly purified streptokinase, plasminogen and plasmin, suggested to me the investigation of the nature of the enzyme or enzymes capable of dissolving fibrin clots. Proteolytic enzymes in general, but more particularly trypsin and chymotrypsin, are known for their ability to digest fibrin clots; certain snake venoms can also do this. The proteolysis of fibrinogen and fibrin by venoms, trypsin or plasmin appears to vary, depending upon which of the enzyme is used. The clotting of fibrinogen is the result of the splitting of an arginine peptide bond in the fibrinogen molecule[1]; this tends to produce a product (fibrin) which is much less susceptible to hydrolysis by trypsin and venoms than the parent substance, fibrinogen[2,3]. Fibrin, on the other hand, is as sensitive to plasmin as is fibrinogen[3].

I wish to report here some attempts made to distinguish a true fibrinolytic enzyme from other proteolytic enzymes which can also dissolve clots.

Fibrinolytic activity as defined by the ability of a substance to dissolve a clot is measured by noting the time required to dissolve a standard fibrin clot under specified conditions. Five-hundredths ml. aliquots of a solution containing various concentrations of the substance to be tested are mixed with 0·1 ml. portions of 0·4 per cent purified bovine fibrinogen[4] in 0·05 *M* veronal buffer of *p*H 7·4 and then to each sample is added 0·05 ml. of thrombin solution containing 10 units of thrombin per ml.; the clot formed in this mixture is kept at 37° C. in an incubator. The time required for lysis (complete dissolution) of the clot is noted in each case. The proteolysis of urea-denatured hæmoglobin has been carried out by the method of Anson[5] and the hydrolysis of synthetic amino-acid esters by the method of Troll *et al.*[6].

Table 1. HYDROLYSIS OF LYSINE ETHYL ESTER BY STREPTOKINASE-ACTIVATED HUMAN SERUM IN THE ABSENCE AND PRESENCE OF AN INHIBITOR, ε-AMINO-CAPROIC ACID

	μM of lysine ethyl ester hydrolysed
Human serum	4·2
Streptokinase + human serum (incubated and then added to the substrate in the presence of amino-caproic acid)	18·6
Streptokinase + human serum + amino-caproic acid (incubated and then added to substrate)	3·4
Streptokinase + amino-caproic acid (incubated and then added to the substrate containing plasminogen (or streptokinase)	10·8

1 ml. of human serum was ten times diluted; to 1 ml. of this diluted serum, 1 ml. of streptokinase containing 1,000 units per ml. and 0·1 ml. of saline or amino-caproic acid are added. 0·2 ml. of this mixture was used in each experiment.

During the course of the experiments it was observed (Table 1) that a preincubated mixture of streptokinase and human plasminogen (that is, proactivator: in human blood the proactivator has been supposed, by some investigators, to be identical to human plasminogen) can hydrolyse synthetic amino-acid esters like lysine ethyl ester in the presence of ε-amino-caproic acid, although a pre-incubated mixture of streptokinase, plasminogen and the inhibitor (ε-amino-caproic acid) cannot do so An appreciable amount of hydrolysis of lysine ethyl ester was, however, noted on the addition of streptokinase (or plasminogen) and ε-amino-caproic acid to a reaction mixture containing the substrate and plasminogen (or streptokinase). Similarly, hydrolysis of this synthetic substrate by plasmin, whether it is prepared from streptokinase-activated human plasminogen (serum) or after treatment of bovine plasma with chloroform or trypsin, is not inhibited by ε-amino-caproic acid. The results indicate that ε-amino-caproic acid can strongly inhibit the reaction of streptokinase with plasminogen to form an activator; once the activation process involving these two substances has occurred ε-amino-caproic acid cannot inhibit the hydrolysis of lysine ethyl ester any further.

As has previously been reported by Sarkar and Devi[7] I have also noted (Table 2) a very strong inhibition of clot lysis by ε-amino-caproic acid if streptokinase-activated human plasminogen (or serum) is used as a lytic agent; inhibition, to a much less extent, is observed if one uses purified plasmin prepared by any of the methods mentioned above. When the concentration of ε-amino-caproic acid is increased five-fold only about 25–30 per cent inhibition is noted. It should also be stated that the ability of streptokinase-activated human plasminogen to dissolve a clot far exceeds the ability of purified plasmin, no matter how the latter is prepared. Trypsin is a poor agent for lysing clot (Table 2). It seems quite likely, therefore, from what has been so far said, that neither plasmin nor trypsin are true fibrinolytic enzymes, although both of these enzymes ultimately can cause clot lysis, by virtue of their proteolytic activity. What I have termed a 'true' fibrinolytic enzyme is one which requires the same

Table 2. EFFECT OF AMINO-CAPROIC ACID ON CLOT LYSIS BY STREPTOKINASE-ACTIVATED HUMAN PLASMINOGEN AND PLASMIN PREPARED BY OTHER METHODS

	No inhibitor	ε-Amino-caproic acid (20 γ)
Streptokinase + human serum	4 min. 10 sec.	7–8 hr.
Plasmin (prepared from streptokinase-activated human serum)	7 min. 45 sec.	40–45 min.
Plasmin (prepared from chloroform-activated bovine serum)	14–15 min.	28–34 min.
Plasmin (prepared from trypsin-activated bovine plasminogen)	14–16 min.	25–30 min.
Trypsin	11–12 min.	15–16 min.

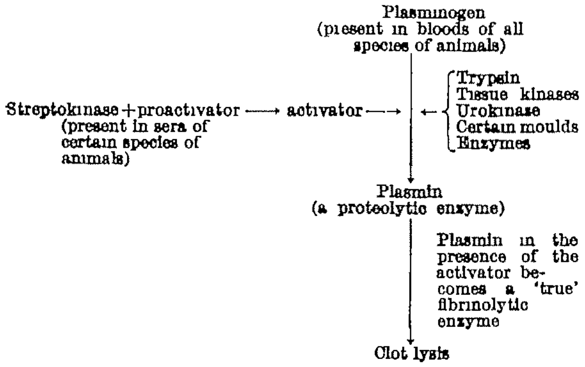

Fig. 1. General mechanism of clot lysis

time for complete dissolution of a fibrin clot and rendering fibrinogen unclottable by thrombin.

What then is the naturally occurring or 'true' fibrinolytic enzyme ? Perhaps there is none. At least a single 'true' fibrinolytic enzyme is not known to-day. Plasmin, once thought to be the enzyme, does not, in fact, occur in the circulating blood. Its inactive precursor plasminogen, present in blood, is transformed to plasmin by an activator, which in turn is formed by an interaction of streptokinase with a proactivator found in the sera of certain species of animals (Fig. 1). Human blood contains this proactivator. It should be noted, however, that some investigators believe proactivator and plasminogen are not identical. Many mould extracts have been reported to be able to dissolve fibrin clot, but it was found by Grey and Sarkar[7] that these extracts dissolve a clot only by virtue of their tryptic activity. Thus it seems that no single substance (enzyme) qualifies at the present time as a 'true' fibrinolytic enzyme. Possibly plasmin, in the presence of the activator, acquires the property of a 'true fibrinolytic enzyme'. The reaction between streptokinase and human plasminogen proceeds with such extreme rapidity that as a rule there is little or no free streptokinase available in the system. This seems to exclude the possibility that streptokinase itself is involved in inducing fibrinolytic activity in plasmin. Whether or not the hypothetical activator of plasminogen itself is causing clot lysis, we do not know; although it looks as though it is the case. Recently, F. B. Ablondi and J. Hagan (personal communication) also expressed such a view.

Recently, Johnson *et al.*[8] demonstrated the inability of plasmin present in the circulating blood to dissolve an artificially formed intravascular blood clot. It was possible, however, to cause dissolution of the clot by introducing further quantities of streptokinase in the blood stream. Thus it appears from these experiments *in vivo* that it is not plasmin alone which dissolves the clot, but rather plasmin in presence of the activator (streptokinase + plasminogen) which does this.

In our attempt to define a 'true' fibrinolytic enzyme, we have mentioned that the ability of such an enzyme to dissolve a clot (clot lysis) and to render fibrinogen unclottable by thrombin should be nearly the same. Trypsin, as reported by Sarkar and Devi[3], needs a much longer time (20 times) to dissolve the clot than it requires to bring about fibrinogenolysis, whereas streptokinase-activated human plasminogen takes the same length of time for both fibrinolysis and fibrinogenolysis (Table 3). Reasons for the difference in the behaviour of these two different types of enzymes should be looked for in the mode of their actions. Enzymes like trypsin dissolve a clot by extensively degrading the protein fibrin. Trypsin also extensively degrades other proteins like denatured hæmoglobin, etc. The method for measuring proteolytic activity of an enzyme depends upon how extensively the enzyme has degraded the protein. In the case of streptokinase-activated human plasminogen it is probable that certain specific linkages holding the fibrin monomers together (to form the gel) are split. This explains why streptokinase-activated human plasminogen is equally active in causing clot lysis and in rendering fibrinogen unclottable by thrombin; whether or not this hypothesis is true is now under further investigation.

Table 3. THE ABILITY OF STREPTOKINASE-ACTIVATED HUMAN SERUM, PURIFIED PLASMINS AND TRYPSIN TO DISSOLVE A CLOT AND RENDER FIBRINOGEN UNCLOTTABLE BY THROMBIN

	Time required for:	
	Fribrinolysis	Fibrinogenolysis
Streptokinase + human serum	4 min. 10 sec.	4 min 30 sec.
Plasmin (prepared from trypsin-activated bovine plasminogen)	14–16 min.	4–5 min.
Trypsin (25 γ)	11–12 min.	30–35 sec.

The work was carried out at the Institute of Laboratories, Boston, Mass., and financed by the Research Division of Jhonson and Jhonson, New Jersey.

A preliminary report of this work appeared in the Proceedings of the Seventh International Congress of Hæmatology held in London, Sept. 7–12, 1959.

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[1] Lorand, L., and Middlebrook, W. R., *Biochem. J.*, **52**, 196 (1952).
[2] Deutsch, H. F., and Dinez, C. R., *J. Biol. Chem.*, **216**, 17 (1955).
[3] Sarkar, N. K., and Devi, Anima, Proc. of Seventh Int. Congr. on Hæmat. (1958).
[4] Laki, K., *Arch. Biochem. Biophys.*, **32**, 317 (1951).
[5] Anson, M. L., *J. Gen. Physiol.*, **22**, 79 (1938).
[6] Troll, W., Sherry, S., and Wachman, J., *J. Biol. Chem.*, **208**, 85 (1954).
[7] Grey, J. B., and Sarkar, N. K. (unpublished work).
[8] Johnson, A. J., McCarty, W. R., and Tillet, W. S., *Surgical Forum*, **9**, 252 (1958).

Lack of Hæmoglobin in a Fish

IN the toad, *Xenopus laevis* Daudin, two specimens have been described completely lacking hæmoglobin[1,2]. I have been able myself to study a teleost fish without hæmoglobin, namely, an adult male of the cichlid *Cichlasoma severum* (Heckel).

The fish, which measured 72 mm. in length, had lived for about two years in a large tank (25 ± 1° C.), was less active than other cichlids of the same litter and during the last few months mostly moved in the surface of the water layer. In this period only little food was taken. The animal became much emaciated, its response to external stimuli almost ceased and ultimately it was killed by decapitation. The animal was smaller than the fishes of the same brood, of which the length varied from 109 to 154 mm.

At autopsy the blood vessels appeared to be white or perfectly colourless except for the chromatophores in their walls. The heart was pale cream and the liver grey. The gills and the kidneys were cream. The gall bladder was enlarged and contained a large quantity of dark green bile pigment. The spleen was enlarged and coloured light brown. The examination of the blood smears showed that hæmoglobin was not identifiable. Erythrocytes were present, however.

The histological investigation showed that the structure of the organs was normal.

On the strength of accumulation of dark green bile pigment, which was also stated in the case of Ewer[2], it may be concluded that the extensive destruction of the hæmoglobin had occurred very recently.

Another point of correspondence with the case of Ewer was the enlarged light brown spleen which, unlike the case of De Graaf[1], was not coloured slightly yellowish-white.

According to De Graaf, his animal without hæmoglobin had been in that condition all its life, with the implication that the growth to maturity in natural conditions is possible without the presence of hæmoglobin. Ewer, however, arrived at a different interpretation, namely, that the total anæmic condition is a pathological state, which may originate after the adult has become mature. My case, in which also the hæmoglobin destruction had only just been completed, is an argument for Ewer's concept.

Survival without hæmoglobin is distinctly possible, which, however, by no means implies that an animal without hæmoglobin can be as active as a normal one. In my specimen the latter state could be clearly distinguished.

According to De Graaf, normal *Xenopus laevis* are capable of functional elimination of at least 80 per cent of their hæmoglobin by carbon monoxide treatment.

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[1] De Graaf, A. R., *J. Exp. Biol.*, **34**, 173 (1957).

[2] Ewer, D. W., *Nature*, **183**, 271 (1959).

Activation of Plasminogen by Staphylokinase

SOME animal plasminogens, which are unaffected by streptokinase alone, are activated by streptokinase and human globulin. Müllertz[1] has postulated the existence of a proactivator in human serum. Staphylokinase, without human globulin, activates a wider range of animal plasminogens[2] and may, therefore, not require proactivator.

Staphylokinase was prepared from the supernatants of fibrinolytic strains of *Staphylococci* shaken for 7 hr. The supernatant was heated at 75° C. for 30 min. Some inert protein was precipitated at *p*H 3·3. After dialysis, staphylokinase was precipitated at *p*H 4·2.

Euglobulin, in the original serum volume, was used as a source of animal plasminogen. Human plasminogen, prepared by the Kline procedure[3], provided proactivator. Plasmin was assayed by the caseinolytic method described by Norman[4], perchloric acid being used instead of trichloracetic acid.

0·2–0·3 ml. of euglobulin was incubated with up to 400 units streptokinase ('Dornokinase', Burroughs Wellcome and Co.) or an equivalent amount of staphylokinase (assayed against human plasminogen), and with or without 3×10^{-3} units human plasminogen. Maximal activity was obtained after 5–10 min. with 20–140 units kinase. Bovine euglobulin did not inhibit the activation of human plasminogen by staphylokinase.

Table 1

	Human	Guinea pig	Rabbit	Bovine
Streptokinase	+	−	−	−
Streptokinase + Proactivator	+	+	+	+
Staphylokinase	+	+	+	−
Staphylokinase + Proactivator	+	+	+	−
Plasminogen (units $\times 10^{-3}$ per ml. serum)	40	57	34	28

The results, which are shown in Table 1, indicate that staphylokinase does not act through proactivator.

A detailed report of this work is being prepared.

I wish to acknowledge the gift of Fraction III of human plasma from E. R. Squibb and Sons, by courtesy of the American National Red Cross.

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[1] Müllertz, S., *Acta Physiol. Scand.*, **38**, Supp. 130 (1956).

[2] Gerheim, E. B., and Ferguson, J. H., *Proc. Soc. Exp. Biol. Med.*, **71**, 261 (1949).

[3] Kline, D., *J. Biol. Chem.*, **204**, 949 (1953).

[4] Norman, P., *J. Exp Med.*, **106**, 423 (1957).

PLANT PHYSIOLOGY

Abnormal Growth and Development in Pea resulting from Exposure to Adverse Conditions during Germination

Kidd and West[1] showed that the amount of growth made by a plant could be determined by the conditions to which the germinating seed was exposed. In their investigations they measured the yield obtained from plants grown from seed soaked in water for varying lengths of time, and they found that in most cases of extended soaking, though the seed germinated more rapidly than seed planted directly in moist soil, the yield obtained from such seedlings was reduced. Their observations of 'physiological predetermination' have been carried further by examining the effect of soaking peas (*Pisum sativum* L.) in water. With this species when seeds are soaked in water for 48 hr. the seedlings that eventually develop may be abnormal (Fig. 1). Normally a small number of seeds, 200, is soaked in a large volume, 500 ml., of tap water kept at 20° C. in darkness. The depth of water covering the seeds was 15 cm., and

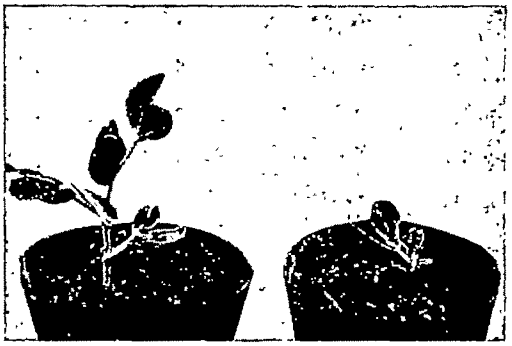

Fig. 1. The plant on the right is a seedling grown from seeds soaked in water for 48 hr. before planting. That on the left is the control planted directly in moist soil

the soaking can best be done in 500-ml. Erlenmeyer flasks.

The effects observed may be related to one or more of the following conditions ; (*a*) A disturbance of the normal metabolism during the first 48 hr. of germination and the maintenance of this abnormal condition during the first stages in development. (*b*) The removal by leaching of an essential water-soluble metabolite. (*c*) The accumulation within the seed of a material which adversely affects growth.

It was thought that peas soaked in stagnant water would respire anaerobically, and therefore it was decided to compare the response of soaked seed with seed which were exposed during the first 48 hr. of germination to an atmosphere containing 20 per cent carbon monoxide. The seeds which were exposed to this atmosphere were kept in darkness, at 20° C., during the period of exposure. In these seeds normal respiration would be blocked since the cytochrome system would be inoperative in this atmosphere. To determine if metabolites were lost, or substances accumulated during soaking, a third sample was kept in running tap-water for 48 hr. At the start of the experiment a fourth sample was planted directly in moist soil in an unheated greenhouse. At the end of treatment the other three samples were sown out in the same bed. After 23 days the plants were harvested, and measurements made immediately on a sample of 30 plants from each treatment (Tables 1 and 2). The number of nodes formed by the largest bud of the first and second nodes (in pea there are usually more than one axillary bud at each node) and the condition of the terminal bud were recorded. If flower primordia were present the number of these was noted, but only when the primordium had recognizable flower parts present ; if no flower buds were present the apex was scored as alive and active, or dead. The remainder of the seedlings produced were stored in 70 per cent alcohol, and from them was obtained the length of the largest axillary bud at the first and second node, and the length of the largest flower present. Gross abnormalities were treated separately. In the batch planted directly into moist soil 161 seedlings were present at harvest and of them 7 were grossly abnormal, while seeds which had been exposed to carbon monoxide produced 171 seedlings with 9 abnormal. Those seeds which were exposed to water produced respectively 129 and 175 seedlings with 34 and 5 abnormals for those soaked in stagnant and running water. The most common type of aberration encountered is suppression of the terminal bud and development of the basal axillary buds. Leaf shape is affected and treated plants are usually slightly chlorotic.

The findings are recorded in Tables 1 and 2.

The results indicate that the normal course of development in pea can be altered by exposing the seed during germination to conditions which affect aerobic respiration. This effect is seen as the removal of apical dominance, in extreme cases by the death of the terminal bud. It appears that the tissues most affected by the treatment are the meristems present in the embryo.

Table 1

	No. of plants with apical buds			Total No. of formed flowers	Average No. of nodes	
	Flowering	Veg.	Dead		Bud No. 1	Bud No. 2
Moist soil	28	1	1	75	2·86	3·76
Running water	28	1	1	49	1·30	3·23
Stagnant water	18	4	8	0	3·50	4·42
20 per cent carbon monoxide	25	1	4	40	1·83	3·24

Table 2

	No. of plants	Average length (relative to moist soil = 100)					
		Bud No. 1		Bud No. 2		Largest flower	
Moist soil	131	5·4	100	9·1	100	5·6	100
Running water	145	5·7	105	9·7	106	5·0	89
Stagnant water	99	6·6	122	11·2	124	3·0	54
20 per cent carbon monoxide	141	6·0	111	9·8	108	2·8	50

This work is being continued to determine the nature of the changes which take place in seeds germinating under the conditions described.

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[1] Kidd, F., and West, C., *Ann. App. Biol.*, 5, 1 (1918).

Nitrate Content of Herbage at Different Manurial Levels

Following on the results reported in *Nature*[1], nitrate nitrogen determinations have recently been made on herbage samples provided by Dr. Rice Williams, of the National Agricultural Advisory Service, from an experiment where sulphate of ammonia had been used at six levels, 0, 2, 4, 6, 8 and 12 cwt. per acre, to swards consisting of a number of different grass mixtures[2]. Nitrogen was applied in mid-April and the herbage was cut six weeks later. The results from one of these mixtures (mixture *B*) are illustrated in Fig. 1. Mixture *B* was seeded down with Italian ryegrass, perennial ryegrass *S*.101, timothy *S*.48, cocksfoot *S*.26, *S*.143, *S*.37 and Danish, red clover *S*.123, alsike, white clover *S*.100 and *S*.184. The experiment was in the form of a randomized block, each level of application having three replicates.

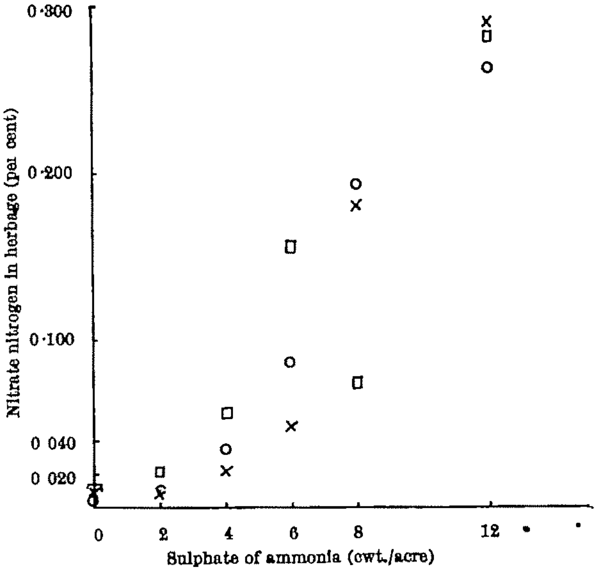

Fig. 1. Relation between levels of nitrogen application and of nitrate nitrogen in herbage. O, ⊡, ×, replications of each treatment

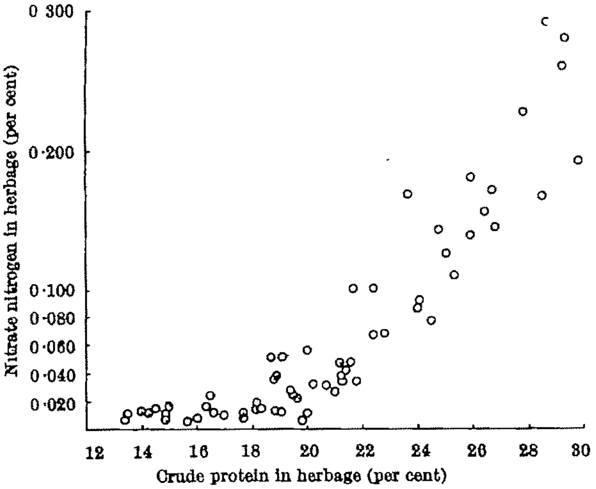

Fig. 2. Relation between percentage crude protein and nitrate nitrogen in herbage

It is seen that nitrogen, applied in the ammonium form, and with a normal interval between application and cutting, has resulted in accumulations of nitrate nitrogen detectable at the 4-cwt. level and considerable at the 8- and 12-cwt. levels.

In Fig. 2, nitrate nitrogen is plotted against crude protein; results from two other grass mixtures in the same experiment are also included. I am indebted to Dr. Rice Williams for the crude protein data.

In amounts greater than 0·07 per cent of the dry matter, expressed as nitrogen, nitrate is reported to be toxic to the animal[3], and amounts greater than 0·22 per cent are reported to be lethal[4]. In the present experiment a crude protein content greater than about 22 per cent would render the material suspect for feeding purposes.

Full details of this investigation are being prepared for publication and will appear elsewhere in due course.

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[1] ap Griffith, G., *Nature*, **182**, 1099 (1958).

[2] Williams, Rice, and Lloyd, J. R., Nat. Agric. Advis. Service, *Quart. Rev.*, **9**, 5, 271 (1952).

[3] Muhrer, M. E., Garner, G. B., Pfander, W. H., and O'Dell, B. L., *J. Anim. Sci.*, **15**, 1291 (1956).

[4] "Chemical Composition of Pasture Plants", Bull. Pa. St. Coll., 480 (1947).

Influence of Hydrogen Ion Concentration and Autoclaving on Gibberellin

In at least two reports the method of sterilization for medium to be used for tissue culture experiments was autoclaving[1,2]. The question of the heat stability of gibberellin is raised. It is generally thought that gibberellin is stable on the basic side of neutrality and at room temperature at neutral or near neutral *p*H, and more labile at elevated temperatures at extreme *p*H values. It has been stated that even weakly acid media will destroy gibberellin and that it is much more stable in neutral and basic solutions (Stowe, B. B., private communication, November 7, 1958).

Several investigators have recently been interested in the heat stability of gibberellin in culture media in the *p*H range 5–6. Experiments have generally been conducted with gibberellin by the sterile-filtration technique. The need for this technique and validity of autoclaving with the medium have been investigated.

Treatments were carried out with autoclaved and unautoclaved stock solutions of gibberellin (supplied by Chas. Pfizer and Co., and consists of gibberellin A and gibberellin X, gibberellic acid) at 100 μgm. per ml. The solutions were buffered to maintain a stable *p*H during autoclaving and treatment of the plants. The buffers at *p*H 1·0 and 3·5 were phthalate–hydrochloride mixtures; *p*H 6·0 was a phthalate–sodium hydroxide mixture; *p*H 10·0 was a boric acid–potassium chloride–sodium hydroxide mixture[3]. Autoclaving was at 20 lb./sq. in. for 15 min.

In all cases the solutions were cooled after autoclaving and placed in the refrigerator for storage before and after use on the plants. During the total length of the experiments (January 17 to December 5, 1959) new stock solutions were made up and new sets of solutions autoclaved for use on the test plants. The solutions, which were re-adjusted to *p*H 6·0, were first autoclaved at the indicated *p*H values and after cooling were adjusted with hydrochloric acid or sodium hydroxide to a *p*H of 6·0 before treatment (0·05 c.c. of the solution) of the plant.

These experiments were set up using the Phinney[4] test for dwarf-1 mutant corn. The plants were treated one week after germination. The dosage was 5γ (0·05 c.c. of 100 γ/ml.) per plant treated only once, at the beginning of the experiment. Recorded readings were taken on the plants two weeks after treatment.

To test whether the *p*H or buffer solutions alone had any effect on growth, the dwarf mutants received likewise 0·05 c.c. of the respective buffer solutions. Measurements of the second internode only were taken at the end of two weeks from treatment. Three plants were used in each pot for each treatment. Results were taken from four to five separate experiments over a 4-month period.

From Table 1, the following observations can be made: (1) all autoclaved solutions possessed less activity than those unautoclaved: (2) the highest activity was from solutions buffered at *p*H values of 6·0 and 3·5, for all treatments, with very little difference between the two; (3) the least activity was from solutions at *p*H 1·0 for all treatments; (4) activity for *p*H 10·0 was intermediate between that at *p*H 3·5 and 6·0, and 1·0; (5) plants treated with buffer alone showed no stimulation whatsoever, being

Table 1. TREATMENT OF DWARF-1 MUTANT CORN SEEDLING, WITH 5 μGM. OF GIBBERELLIN AT VARIOUS BUFFERED *p*H VALUES, AFTER AUTOCLAVING OR NOT AUTOCLAVING

Measurement of the second internode in centimetres after two weeks treatment; numbers in brackets are number of plants included to calculate average

Treatment	*p*H of buffer			
	1·0	3·5	6·0	10·0
Unautoclaved	2·5±1·2* (12)	6·8±2·0 (15)	6·6±1·4 (12)	5·2±1·4 (11)
Autoclaved	1·3±0·4 (10)	4·6±1·5 (14)	4·9±1·8 (12)	3·3±1·1 (12)
Autoclaved, readjusted to *p*H 6·0 after autoclaving	1·8±0·6 (10)	4·6±0·9 (13)	4·5±1·3 (15)	3·3±1·4 (11)
*p*H control	1·0 (3)	1·2 (3)	1·5 (3)	1·0 (2)
Normals	5·5±2·3 (22)			
Dwarf controls	1·2±0·4 (12)			

* Standard error of the mean

the same as the dwarf controls; (6) solutions initially autoclaved in the various buffers and afterwards adjusted to a common *p*H of 6·0 were uninfluenced by this post-autoclaving treatment; (7) all gibberellin solutions, regardless of type of treatment, showed greater activity than solutions containing no gibberellin (*p*H control plants), and caused greater growth of the dwarfs than the untreated (dwarf) controls; (8) the unautoclaved gibberellin solutions at *p*H 3·5 and 6·0 showed greater activity than the normal, non-dwarf plants; (9) the reduction in activity of gibberellin due to autoclaving was in no case greater than 50 per cent (for *p*H 1·0) and for solutions at *p*H 3·5 and 6·0 it was roughly 33 and 25 per cent, respectively.

From these observations one may draw the following conclusions: (1) There is significant (statistical) loss of activity caused by autoclaving of gibberellin, but this loss is not so significant biologically (see explanation below); (2) the loss of activity is between one-third and one-fourth of the original activity at the optimum *p*H value for most culture solutions employing gibberellin, especially sterile tissue culture; (3) based on the experiments with *p*H controls and the re-adjusted autoclaved solutions, the *p*H factor itself does not influence these plant responses directly; the effect of heat and *p*H is rather on the gibberellin molecule itself, and subsequent influence of the latter on plant response.

The general over-all conclusion is that the autoclaving of gibberellin at the normal *p*H of culture media is not biologically significantly detrimental or destructive to the gibberellin content in the medium. From standard curves of biological activity, as well as response in plant tissue culture, the loss of as little as one-third to one-fourth, or even more, of the activity of gibberellin at any single concentration is never very significant, since most experiments employ the logarithm of the concentration of growth factors instead of linear units. Autoclaving of other organic growth factors and auxins, especially indoleacetic acid, would show approximately similar losses. Therefore, it is generally safe to use the autoclaving technique in the usual experiments employing gibberellin in plant tissue culture, especially where the amounts employed are greater than 1 p.p.m.

Since these conclusions are based on one of the only reliable methods for the assay of gibberellin at present—the bioassay technique, limited to the assay with Dwarf-1 mutant corn in this instance—it is not possible to speculate what the active compound(s) is (are). It is specifically adapted to each bioassay test. Further work, both biological and chemical, is required to identify the active principle.

I wish to thank Chas. Pfizer and Co., Inc., for supplying the gibberellin, and the National Science Foundation for supporting the investigation. Mr. Clarence Saulsberry assisted in the experimental work; Dr. B. O. Phinney supplied the Dwarf-1 mutant seeds.

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[1] Schroeder, C. A., and Spector, C., *Science*, **126**, 701 (1957).

[2] Henderson, James H. M., *Nature*, **182**, 880 (1958).

[3] Koch, F. G., and Hanke, M. E., "Practical Methods in Biochemistry", 99 (Williams and Williams, 1948).

[4] Phinney, Bernard O., *Proc. U.S. Nat. Acad. Sci.*, **42**, 185 (1956).

RADIOBIOLOGY

Effectiveness of Dietary Substances in reducing the Retention of Chronically Ingested Radiostrontium; Studies with Tannin

THERE has been considerable interest in the testing of substances that may reduce the body burden of chronically ingested radiostrontium. The present work, using tannin as an example, emphasizes the caution that must be used in the interpretation of single-dose studies, and also describes a relatively short-term procedure which appears to be practical.

In a recent report, Ukai *et al.*[1] have shown that the skeletal retention of a single dose of strontium-90 could be reduced if followed immediately by the oral administration of a 1 or 2 per cent tannin solution. It has been suggested from these results that consumers of tannin (in tea) would absorb and retain less fall-out strontium-90 than otherwise. Such interpretations of data from single-dose studies applied to situations of normal strontium-90 ingestion can be especially misleading. First, strontium-90 is ingested continuously as a contaminant in most food products; secondly, strontium-90 is consumed simultaneously with dietary calcium and other constituents of the diet. Under these conditions, a complexing agent may yield an entirely different effect from that when it is given as a single dose with carrier-free radiostrontium. In fact, our previous experience has shown that substances which complex or chelate strontium and calcium, for example, citrate, phytate, sodium ethylenediaminetetraacetate, were without effect in reducing the body burden of continuously ingested radiostrontium[2]. In view of this prior knowledge, it was felt necessary to determine the influence of tannin on radiostrontium and radiocalcium retention in the situation where (*a*) the radiostrontium is continuously ingested over a period of days, (*b*) the tannin is consumed over the same time period, and (*c*) the normal constituents of the diet are also present.

Twenty-four rats were divided into four groups of six rats each and placed in individual cages. Drinking-water labelled with essentially carrier-free calcium-45 and strontium-85 was made available in graduated drinking tubes; the levels of calcium-45 and strontium-85 were 0·1 and 0·01 μc. per ml., respectively. At the same time, the rats were given a complete semi-synthetic diet containing either 0, 1, 2 or 4 per cent tannin (Nutritional Biochemicals, Inc.); the level of calcium in these diets was about 0·48 per cent. The rats were allowed to ingest the 'spiked' water and diet *ad libitum* over an experimental period of ten days. The amount of water and diet consumed was measured. At ten days, the rats were killed and retention of calcium-45 and strontium-85 in the femur was determined by usual radioassay procedures. The results are given as 'per cent of total ingested activity'. It should be pointed out that previous studies have shown that radiostrontium and radiocalcium ingested continuously in drinking water equilibrate with, and act similarly to, these elements ingested in the diet[3].

The results are presented in Table 1. It may be seen that the presence of tannin in the diet at the higher levels appreciably decreased the growth-rate of the experimental animals although feed consump-

Table 1. INFLUENCE OF TANNIN ON THE FEMUR RETENTION OF CONTINUOUSLY INGESTED CALCIUM-45 AND STRONTIUM-85

Group	Concentration of tannin in diet (per cent)	Body-weight*			Feed intake* (gm.)	Femur retention of radionuclides†		OR‡
		Initial (gm.)	Final (gm.)	Gain (gm.)		^{45}Ca (per cent ingested dose)	^{85}Sr (per cent ingested dose)	
1	0	235	261	26	143	2·3 ± 0·3	0·46 ± 0·03	0·20 ± 0·01
2	1	237	257	20	174	2·3 ± 0·2	0·58 ± 0·08	0·25 ± 0·02
3	2	243	262	19	159	2·1 ± 0·1	0·66 ± 0·06	0·30 ± 0·02
4	4	239	242	3	159	2·0 ± 0·1	0·64 ± 0·03	0·32 ± 0·04

* Body-weight and feed intake given over 10-day period.

† Values represent mean ± standard error of the mean of six animals per group.

‡ Observed ratio (OR) = ^{85}Sr/^{45}Ca in femur ÷ ^{85}Sr/^{45}Ca in diet.

tion was not reduced. In regard to radionuclide retention, a slight decrease in radiocalcium retention by the rat femur was noted in the tannin groups in contrast to the controls. However, in the presence of 2–4 per cent dietary tannin, the strontium-85 content of the rat femurs was significantly increased, causing an increase of the strontium/calcium observed ratio. An increased strontium/calcium ratio had similarly been observed in rats fed a liquid milk diet containing sodium ethylenediaminetetraacetate. Since tannin and ethylenediaminetetraacetate possess the common property of binding calcium ions to a somewhat greater degree than strontium ions, the present results may possibly be explained as follows. Radiostrontium previously complexed with tannin in the gut could be replaced by stable calcium and, therefore, become available for absorption. Parental tannin would act similarly, that is, releasing complexed strontium-85 when replaced by plasma calcium.

The above results are not in direct conflict with those of Ukai *et al.*[1] because entirely different conditions were employed in the two studies. Any disagreement would be in the extrapolation of single-dose data to the chronic situation in which strontium-90 is ingested with the diet at each meal. When this chronic situation is experimentally simulated as in the present study, it was observed that tannin at various levels in the diet had *no* beneficial effect in reducing the body burden of radiostrontium in the rat. Although single-dose studies of the type referred to may have usefulness in readily assessing the potential value of any test substance, it is recommended that promising compounds should be subjected to a chronic ingestion test before extrapolations are made to the chronic situation. It is further suggested that the influence of the test substance should be simultaneously determined on radiocalcium retention, since interpretations are facilitated and important information of possible effects on an essential nutrient is thereby provided.

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[1] Ukai, T., Hayashi, E., and Saito, S., Abst. Amer. Pharmaceut. Assoc., Sci. Sec., Cincinnati, Ohio (Aug. 19, 1959).

[2] Comar, C. L., Russell, R. Scott, and Wasserman, R. H., *Science*, **126**, 485 (1957).

[3] Wasserman, R. H., Comar, C. L., and Papadopoulou, D., *Science*, **126**, 1180 (1957).

Early Injury of Chromoexcretion after Total Irradiation of Rats

IT is claimed by all available literature that damage occurs to the function of the liver after whole-body irradiation by doses, approaching *LD*100, from as late as the stage of development of the secondary post-radiation changes[1,2]. Some biochemical results, however, support the opinion that the liver cell should be damaged in its function at the earliest stage of an acute irradiation disturbance[3-5]. To prove the early functional lesion we carried out experiments with bengal rose tagged with radioactive iodine-131.

Experiments were performed on 50 male rats of the Wistar strain averaging 200 gm. in weight. Twenty-six rats were irradiated with a dose of 600 r. by the deep X-ray apparatus of 200 kV., 15 m.amp., filtration 3 mm. aluminium at a distance of 75 cm. and 32 r./min. The remaining 24 rats were used as a control group. Tests were accomplished in 4 groups, of 6 and 8 rats respectively, the controlled and irradiated ones, 2, 6 and 24 hr., and 14 days after the irradiation. At the given intervals we applied to the tail vein of the rats 5 μc. bengal rose–iodine-131 in 1 ml. of isotonic solution[6]. 10 min. after the injection the rats were killed, the liver was taken out and homogenized separately.

Gamma activity of the homogenates was determined by the scintillation method. The activities of the control-group homogenates were taken as 100 per cent and the activities of the irradiated groups were expressed as a percentage of the control-group activity.

In the group tested 2 hr. after the irradiation the uptake value of 81 per cent, as compared with 100 per cent of the control group, was still above the 5 per cent limit of significance. The further group, tested 6 hr. after irradiation, has its uptake lowered to 83 per cent of limit of error below 1 per cent. The group killed 24 hr. after irradiation has its uptake lowered to 76 per cent with 0·5 per cent limit error, and that killed on the fourteenth day after irradiation exhibits lowering to 74 per cent with ·1 per cent significance.

Uptake of bengal rose is a specific property of polygonal liver cells. Radioactive bengal rose permits differentiation of the great damage done to the blood—or bile pole of the liver cell. In preliminary experiments, during investigations for the most suitable moment for homogenization after injection, we ascertained the maximum difference in the tenth minute after the application of colouring matter, that is, at the uptake maximum. This fact is an indication of considerable damage caused to the blood pole of the liver cell. The question of this mechanism is still open, and is the subject of further investigation.

The possibility of distributive volume changes of bengal rose is not likely to make itself felt to any extent in these results. This is due to the fact that distributive volume for bengal rose is practically equal to the plasma volume, and there is no literature known to us suggesting that considerable changes occur in the plasma volume in the early stages after the irradiation of the whole body.

We believe that the significant lowering of uptake maximum in the sixth hour after irradiation explains the early injury to the chromo-excretional function in rats after whole-body irradiation by lethal doses of 600 r. using bengal rose–iodine-131.

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[1] Du 'Bois, K. P., and Petersen, D. F., "Ann. Rev. Nucl. Sci.", 1, 351 (1954).

[2] Thomson, J. F., and Mikuta, E. T., *Proc. Soc. Exp. Biol. Med.*, 86, 487 (1954).

[3] Hagen, N., Koch, R., and Langendorff, H., *Fortsch. Rontgen.*, 84, 604 (1956).

[4] Belovinceva, M. F., and Speranskaja, J. H., "Problem of Significance of Function State of Liver in Evolution and Therapy of Irradiation Disease", All Union Conference on Use of Isotopes in Irradiation (Moscow, 1957). (In Russian.)

[5] Skalka, M., *Czech. Biol.*, 7, 424 (1958).

[6] Liebster, J., and Andrysek, O., *Nature*, 184, 914 (1959).

BIOLOGY

Thermal Barrier to the Infectivity of *Entamœba invadens* in Snakes

THE optimum temperature for the *in vitro* growth of *Entamœba invadens* has been determined by several investigators to be between 20 and 30° C.[1,2]. I found that the maximum temperature at which *E. invadens* could be grown in developing chicken embryos was 30° C.[3]. The following account represents the results of a preliminary investigation on the ability of *E. invadens* to establish itself in snakes kept at temperatures above normal.

Three adult grass snakes, *Thamnophis sirtalis*, free of parasitic Amœbæ, were used in each of the two experiments. All the snakes were inoculated *per os* with equal doses of a heavy suspension of trophozoites and cysts of the *I.P.*2 strain of *E. invadens*[4], taken from polyxenic cultures.

In the first experiment snakes 1 and 2 were placed in individual jars in a hot room, in which the temperature fluctuated between 34° and 37° C. Snake 3 was kept in the laboratory, the room temperature varying from 20° to 23° C. During the course of the experiment the snakes' fæces were examined for the presence of Amœbæ and of blood, which is an indication of intestinal ulceration produced by the Amœbæ. The snakes' rectal temperature was recorded at the same time by means of a very fine thermometer. Table 1 represents the findings in Exp. 1.

Table 1

	Days after inoculation	Room temperature (deg. C.)	Rectal temperature (deg. C.)	Microscopical examination of fæces
Snake 1	1	35·5	34·5	Negative
	2	37	32	Negative
	4	34	34	R.B.C., no Amœbæ
	5	35·5	34·5	R.B.C., no Amœbæ
	6	35	33·5	No Amœbæ
	7	34·5	34	No Amœbæ
	8	Died. No pathological changes and no Amœbæ seen in internal organs.		
Snake 2	1	35·5	34·5	Negative
	2	37	32	Negative
	4	34	34	R.B.C., no Amœbæ
	5	35·5	34·5	R.B.C., Amœbæ present
	6	35	34·5	Few Amœbæ
	7	34·5	34·5	Few Amœbæ
	8	Died. No pathological changes and no Amœbæ seen in internal organs.		
Snake 3	1	21	20	Negative
	2	22	22	Negative
	4	20	20	Negative
	5	20·5	19·5	Blood and Amœbæ
	6	21·5	21	Blood and Amœbæ
	7	23	25	Blood, no Amœbæ
	8	Killed. Large intestine hæmorrhagic and ulcerated. Amœbæ recovered in cultures made from the intestine and liver.		

Three other snakes were similarly inoculated in the second experiment; Nos. 1 and 2 were placed in separate jars in the hot room and No. 3 was left in the laboratory.

Snake 1 died two days after inoculation. Upon autopsy Amœbæ were not seen in its intestine or liver, but a culture made from the intestinal contents became positive for *E. invadens*.

On the seventh day after inoculation the rectal temperature of snake 2 was 33° C., while that of the room was 34·5° C. There were no living Amœbæ in its fæces on that day, but many cells resembling dead Amœbæ were observed. A sample of fæces was taken from the same snake on the fifteenth day after inoculation, but no Amœbæ were seen in it, nor did the cultures made from the fæces become positive for Amœbæ. Snake 2 was found dead on the seventeenth day after inoculation. There were no pathological changes in its intestine or liver. Cultures made from these organs did not become positive for Amœbæ. It was also noted that the flagellates, which were very numerous in the intestine of this snake at the beginning of the experiment, were all gone when autopsy was performed.

Snake 3, which was kept in the laboratory, developed typical amœbiasis about a week after inoculation and was killed on the tenth day. Its large intestine was heavily ulcerated, and *E. invadens* were seen in the intestine and the liver.

These preliminary experiments have shown that *E. invadens* is unable to establish infections in snakes at high temperatures, although, as seen in snake 2 in Exp. 1 and in snake 1 in Exp. 2, the Amœbæ survived in the intestine for a few days. It is also interesting that the flagellates, often present in snakes' intestines, apparently could not survive at high temperatures. The snakes kept at high temperatures died within relatively short periods of time, although their death was not due to the amœbic infection.

It is planned to repeat these experiments, using strains of *E. invadens* which have been adapted to *in vitro* growth at 35° C.

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[1] Rodhain, J., *C.R. Soc. Biol., Paris*, 117, 1195 (1934).

[2] Gelman, Q. M., and Ratcliffe, H. L., *Parasitol.*, 28, 208 (1936).

[3] Meerovitch, E., *Canad. J. Microbiol.*, 2, 1 (1956).

[4] Meerovitch, E., *Canad. J. Zool.*, 36, 423 (1958).

Variations in the Plasma-Level of Chloride in Hatchery-reared Yearling Atlantic Salmon during Parr–Smolt Transformation and following Transfer into Sea-Water

The successful adaptation of juvenile Atlantic salmon to sea-water requires a prior physiological 'metamorphosis'—parr–smolt transformation[1]. The occurrence of this change appears to depend in part upon age, and in part upon the complex of factors controlling the growth-rate of the parr[2]. In addition to external evidences of parr–smolt transformation such as loss of parr marks, decrease in condition factor and deposition of guanine there must also be some variation in biochemical systems associated with the regulation of body electrolyte-levels relative to those of the environment. Evidence for such changes may be seen in the changing plasma chloride-levels of two migratory salmonids, *Oncorhynchus masou* and *Salmo gairdneri*, during transition from parr to smolt stages[3]. Several investigators have also reported marked development of Keys–Willmer cells, units thought to have an ion secretory role, in a number of species of salmon prior to downstream migration[4]. Fontaine, however, could find no significant variation in blood chloride-levels of smolting Atlantic salmon, although a decrease did occur in tissue concentration[5].

During the course of an investigation of factors influencing the adaptability of hatchery-reared yearling Atlantic salmon to sea-water it was possible to observe plasma chloride-levels in fish maintained in freshwater over a five-month period. The animals were obtained from the Grand Lake Fish Culture Station, Nova Scotia, and were held in recirculating aquaria at 11° C. in the laboratory. Analysis of chloride content was carried out by the mercurimetric titration procedure on 50 and 100 lambda-plasma samples. Samples were obtained by exposed cardiac puncture from anæsthetized animals.

Fig. 1 summarizes results obtained on the 45 animals sampled. These were assessed at the time of sampling as parr (parr marks distinct, no silvering), silvery parr (parr marks more or less distinct, some silvering) or smolt (parr marks absent, marked silvering). In small parr (6–15 gm.) chloride-levels were relatively size-stable and ranged from 125 to 139 m*M*/l. In somewhat larger parr (15–26 gm.) plasma chloride decreased markedly with increase in weight. In silvery parr (17–31 gm.), on the other hand, chloride concentrations increased with increases in weight to a maximum of 145–150 m*M*/l. A slight decrease from this level was observed in the smallest smolt sampled, but values for larger animals were relatively constant at between 130 and 140 m*M*/l.

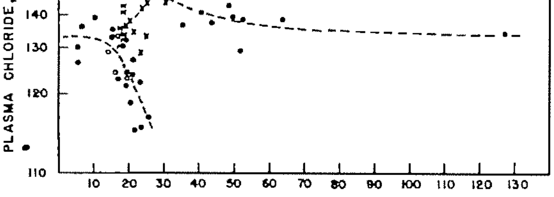

Fig. 1. Variation in plasma chloride concentration with weight and stage of development in freshwater-adapted Atlantic salmon

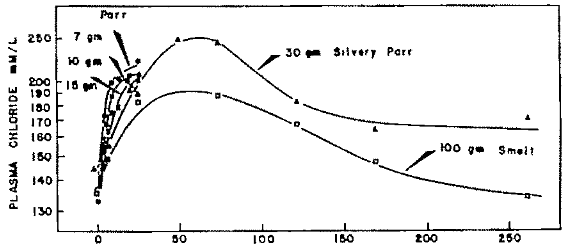

Fig. 2. Plasma chloride concentrations of parr (7, 10, 15 gm.), silvery parr (30 gm.) and smolt (100 gm.) following transfer from freshwater into sea-water (chloride concentration: 400–410 m*M*/l.)

From the observations it is inferred that the initiation of parr–smolt transformation is accompanied by alterations in the electrolyte metabolism of the parr which lead to a loss in regulatory ability. Salt balance in freshwater fishes involves the precise adjustment of uptake processes (dietary gain, active absorption) with processes leading to loss or dilution (urinary output, outward diffusion). A predominance of the latter group of factors is indicated in late parr, while a reversal of this effect is apparent in silvery parr. The results suggest, as well, that the process of smoltification may be well under way before any obvious indications of its occurrence are present, since chloride imbalance was present in parr displaying distinct parr marks and the absence of any silvering.

Variation in patterns of endocrine activity may account for the distinct decrease in regulatory efficiency which is apparent in the early stages of parr–smolt transformation. Data assembled by Pickford and Atz and by Hoar[6] suggest that increased pituitary, thyroid and adrenal cortical activity accompany smoltification. Of particular interest are the demonstrations by Hoar[4] of a relationship between thyrotropic hormone and the development of Keys–Willmer cells in Atlantic salmon parr, and by Fontaine and Hatey[7] of marked increases in the blood-levels of 17-hydroxycorticosteroids in the smolt stages of the same species.

Chloride-levels of fish transferred from freshwater directly into sea-water (400–410 m*M*/l.) are recorded in Fig. 2. Values were interpolated from regression equations relating plasma chloride to body-weight in groups of 15–20 fish.

Small parr (7, 10, 15 gm.) took up chloride rapidly and none survived for longer than 26 hr. Smaller parr took up chloride more rapidly than did larger animals, and died earlier. This observation is consistent with those made earlier by Huntsman and Hoar[1] on the same species.

Animals in the process of transformation appeared to be developing the capacity to become adapted to sea-water. Increases in chloride-level were, however, more marked than those observed in smolt, and the silvery parr required a longer time to return to freshwater chloride-levels than did completely transformed fish. The process of adaptation to sea-water required about ten days in fully transformed smolt.

In general, this study tends to support the conclusions of Koch, Evans and Bergstrom[8] based upon sodium regulation in Atlantic salmon parr and smolt. A loss of regulatory capacity occurs while the fish become transformed from the freshwater-adapted parr stage to the potentially marine smolt stage, while adaptation to sea-water appears to be dependent upon both size and stage.

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[1] Fage, L., *Bull. Inst. Oceanog., Monaco*, **225**, 1 (1912). Huntsman, A. G., and Hoar, W. S., *J. Fish. Res. Bd. Canada*, **4**, 409 (1939). Parry, G., *Nature*, **181**, 1218 (1958).

[2] Elson, P. F., *Canad. Fish. Cult.*, **21**, 1 (1957).

[3] Kubo, T., *Bull. Fact. Fish., Hokkaido Univ.*, **6**, 201 (1955). Houston, A. H., *Canad. J. Zool.*, **37**, 729 (1959).

[4] Hoar, W. S., *Univ. Toronto Biol. Ser.*, **59**, 1 (1951). Nishida, H., *Sci. Rep. Hokkaido Fish. Hatch.*, **8**, 83 (1953).

[5] Fontaine, M., *C.R. Acad. Sci., Paris*, **232**, 2477 (1951).

[6] Pickford, G. E., and Atz, J. W., "The Physiology of the Pituitary Gland of Fishes" (N.Y. Zool. Soc., 1957). Hoar, W. S., "Endocrine Organs", Chap. 6 in the "Physiology of Fishes", 1, ed. by Brown, M. E. (Academic Press, 1957).

[7] Fontaine, M., and Hatey, J., *C.R. Acad. Sci., Paris*, **239**, 319 (1954).

[8] Koch, H. J., Evans, J. C., and Bergstrom, E., *Nature*, **184**, 283 (1959).

Study of Life-cycle of *Schizosaccharomyces octosporus* by means of Ultra-thin Sectioning and Electron Microscopy

CYTOLOGICAL studies of the yeast of the genus *Schizosaccharomyces* have been few and limited primarily to observations by means of light microscopy[1-3]. Although these studies reveal the nature of some of the general cytological changes which occur during various phases of the life-cycle of these yeasts, detailed studies are lacking. Some of the conflicting views and limited observations of these investigators may be attributed at least in part to the resolution limit of the light microscope. A study of the nature and sequence of the cytological changes which occur during cell division, conjugation, ascosporogenesis, and germination was therefore carried out by means of ultra-thin sectioning and electron microscopy in order to obtain a more detailed and accurate description of the cytological changes which occur. Various cytochemical methods were also employed in order to correlate structures observed in electron micrographs with structures observable with the light microscope. *Schizosaccharomyces octosporus*, N.R.R L. Y-854, obtained from Dr. L. J. Wickerham, was selected for study. The techniques used for specimen preparation have been described previously[4].

Fig. 1,*a–i* illustrate the nature and sequence of the cytological changes accompanying cell division. It was observed that karyokinesis occurs well in advance of cytokinesis and that transverse cell-wall formation occurs by the annular centripetal growth of an inner lateral portion of the cell wall. The cytoplasmic membrane appears to remain in close proximity to the cell wall during cytokinesis. After formation the transverse cell wall increases in thickness and electron-dense membrane-like structures are formed. The transverse wall then splits as shown, the transverse membranes serving initially as the outer portion of the cell wall. These membranes may be analogous to the splitting plate observed in dividing cells of *Histoplasma*[5]. Nuclear division appears to occur by a process of elongation and constriction, further observation of chromatin behaviour being beyond the scope of the techniques employed. The nuclear membrane appears to remain intact throughout all the observed stages of nuclear and cytoplasmic division.

The conjugation process (Fig. 1,*j–o*) appears to be initiated by the attachment of cells in pairs by a portion of the cell wall. The central portion of the connecting wall disintegrates, thereby forming a short, narrow conjugation canal which afterwards increases both in length and width. During this stage the nuclei migrate toward one another and fuse within the canal. The nuclear membrane remains intact except in the region of nuclear fusion. Vacuoles are formed in the majority of cells during or immediately after fusion nucleus formation. The fusion nucleus (presumably diploid) then undergoes a series of divisions resulting in the formation of haploid nuclei (Fig. 1,*p–u*).

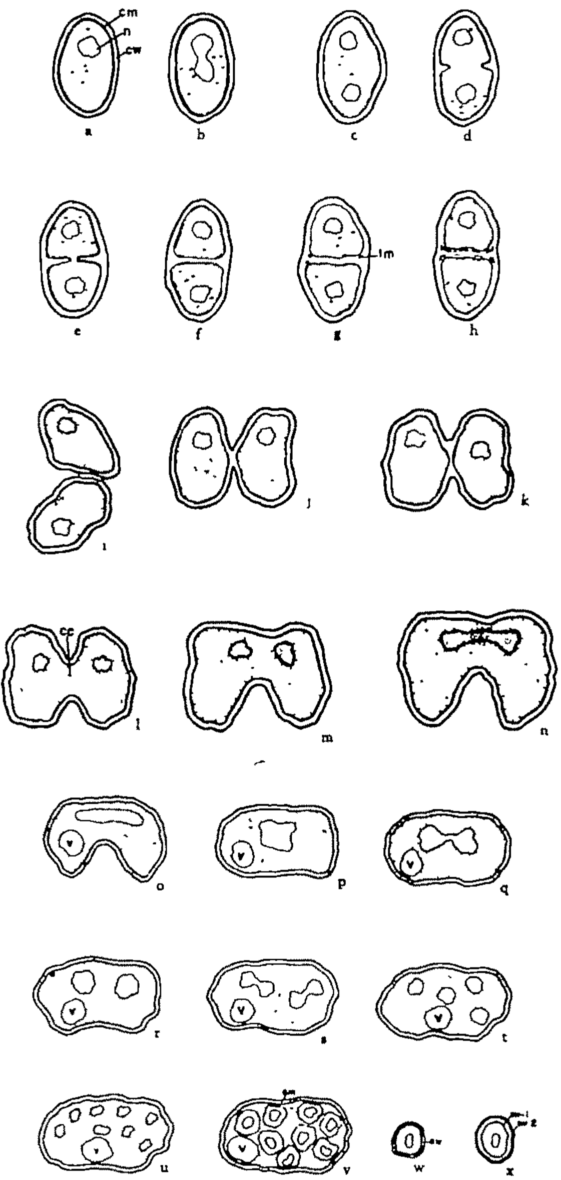

Fig. 1. Life-cycle of *S. octosporus*. *a–i*, Cell division; *j–v*, conjugation, nuclear division and sporulation; *w*, developing ascospore; *x*, appearance of a mature ascospore prior to germination. *AM*, Ascospore membrane; *AW*, ascospore wall; *CC*, conjugation canal; *CM*, cell membrane; *CW*, cell wall; *N*, nucleus; *SW*-1, inner, non-electron dense portion of the ascospore wall; *SW*-2, outer, electron-dense portion of the ascospore wall; *TM*, transverse membrane; *V*, vacuole

Sporulation (Fig. 1,*v*) is initiated by the formation of a double ascospore membrane around each nucleus. The inner portion of the ascospore membrane serves as the structural precursor of the cytoplasmic membrane. The ascospore wall then develops around the outer portion of the ascospore membrane (Fig. 1,*w*). Mature and germinating ascospores are surrounded by two spore coats (Fig. 1,*x*), the inner one giving rise to the vegetative cell wall during germination. Germination is initiated by swelling and elongation of the ascospores with subsequent disruption of the outer spore coat. Vegetative cell division is then initiated.

The results of this study generally support and extend considerably the previous observations on cell division, sporulation, and germination in *S. octosporus*[1-3]. The electron micrographs from which the illustrations are drawn, and a more detailed description of the results will be presented elsewhere.

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[1] Guilliermond, A., "The Yeasts" (John Wiley and Sons, New York, 1920).

[2] Widra, A, and DeLamater, E. D, *Amer J. Bot*, **42**, 423 (1955).

[3] Knaysi, G, *J Bacteriol.*, **41**, 141 (1940).

[4] Conti, S. F., and Naylor, H. B, *Bact. Proc.*, 50–51 (1959).

[5] Edwards, M. R, *et al.*, *J Gen. Microbiol*, **20**, 496 (1959).

BACTERIOLOGY

Antigens of *Azotobacter*

Previous attempts at studying the antigens of *Azotobacter* have met with only limited success. Azo and Yoshida in a brief report[1] described complement-fixation by *Azotobacter* cells in the presence of specific antisera but gave few experimental details. Jensen and Petersen[2] obtained an agglutinating antiserum for *Azotobacter chroococcum*. Recently, Petersen[3] studied the agglutinogens of *Az. chroococcum*, *Az. vinelandii* and *Az. agilis*. *Az. chroococcum* and *Az. vinelandii* had antigens in common and showed serological affinities to *Beijerinckia* sp. *Az. agilis* appeared to be antigenically distinct Eisenstark *et al.*[4], on the other hand, using apparently adequate technical methods, failed to produce antisera to *Az. agilis* or "numerous other azotobacter species". Heidelberger (quoted in Lawson and Stacey[5]) showed *Az. chroococcum* polysaccharide to be antigenic.

A knowledge of the antigenic structure of *Azotobacter* could be of value in two ways: the genus presents taxonomic problems which may be clarified by a serological study[3] and the large cells are eminently suitable subjects with which to study the distribution of antigens within the cells. Methods are available for the fractionation of these organisms into their structural components[6]

In the present investigation cultures were grown on glucose, nitrogen-free, agar at 30° for two days and harvested into sterile saline. Antisera to *Az. agilis* (V J.) were prepared by injecting freshly prepared untreated cell suspensions, adjusted to an opacity equal to that of Brown's tube No. 8, intravenously into rabbits. A series of injections consisting of 0·5, 1·0, 2 0, 2·0, 2 0 and 2·0 ml. at 3-day intervals was followed by a rest period of 1 month, after which three further injections of 1·0, 2 0 and 2 0 ml. were given. Antisera were stored without preservative at −20°.

Table 1. Distribution of *Azotobacter agilis* Antigens among Other *Azotobacter* Species

Species (strain)	No of antigens detected	Known antigens (referred to a standard gel diffusion plate—antigen/antibody lines numbered from the antiserum reservoir outwards)				
Az. agilis (V J.)	5	1	2	3	4	5 (complex)
Az. agilis (4 4)	5	1	2		4	5 (partial)
Az. insigne (V.J.5)	4		2		4	
Az. macrocytogenes (0)	4		2		4	
Az. chroococcum (222)	1				4	
Az. beijerinckii (L.H 1)	2				4	
Az. vinelandii (0)	3				4	

Antigen–antibody reactions were studied by the Ouchterlony gel diffusion technique. Cells from 2-day old agar cultures were harvested into sterile saline containing 1 in 10,000 thiomersalate and deposited by centrifuging. The deposit was resuspended in a minimal amount of the same medium to give a thick paste of cells which could just be handled conveniently with a Pasteur pipette. The resulting cell suspensions were used as antigen preparations. Sensitivity and resolution of lines were improved by using thin layers of diffusion medium, and after several trials 0·5 ml. amounts of a 1·7 per cent agar/saline base containing 1 in 10,000 thiomersalate in 3½-in. Petri dishes were chosen as the conditions giving the best results. Antigen suspensions and antisera were accommodated in cork-borer holes, and the Petri dishes were sealed into small plastic bags and incubated at 30°. Although precipitation lines could be seen after 24 hr., the full complement of lines required up to two weeks to develop—presumably some antigens are only slowly released from the cells under these conditions. Fully developed plates were preserved by drying, stained with naphthalene black, and photographed against a yellow safelight screen.

Fig. 1 shows a typical result obtained by allowing antigens from six species of *Azotobacter* to diffuse against *Az. agilis* (V.J.) antiserum. *Az. agilis* (V.J.) has at least five antigens which may be numbered from the antiserum reservoir outwards for reference. One of the antigens (No. 4) has been found in all strains of *Azotobacter* so far studied. The fifth antigen, giving the heaviest line on the plates, is clearly complex and is occasionally seen to consist of numerous components (Fig 2) some of which are present in other strains of *Az. agilis* (Fig. 3). The details of the distribution by species of all the antigens

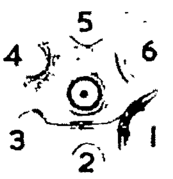

Fig. 1

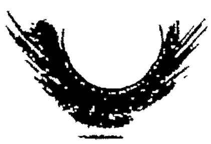
Fig. 2

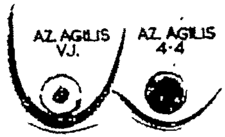

Fig. 3

Fig 1. Gel diffusion plate. Antiserum to *Azotobacter agilis* (V.J) in the centre. Antigens: 1, *Az. agilis* (V J.); 2, *Az. insigne* (V.J.5); 3, *Az. vinelandii* (0), 4, *Az. beijerinckii* (L.H.1); 5, *Az. chroococcum* (222), 6, *Az macrocytogenes* (0)

Fig 2. Gel diffusion plate. *Az. agilis* (V.J) antigen diffusing against the homologous antiserum. Photograph showing the complexity of antigen No. 5

Fig 3. Gel diffusion plate showing the partial sharing of the complex antigen No. 5 between two strains of *Az. agilis*. Antigen suspensions diffusing against *Az. agilis* (V.J) antiserum

are not yet completely worked out, but Table 1 summarizes the distribution as it is understood at the moment. The suggestion of a close relationship between *Az. agilis*, *Az. insigne* and *Az. macrocytogenes* is of great taxonomic interest. The closeness of *Az. agilis* and *Az. insigne* is well known[7], but the relationships of *Az. macrocytogenes* are not fully understood, although Johnstone[8] has shown that it shares with *Az. agilis* the ability to produce a water-soluble pigment giving a white fluorescence in ultra-violet light. I have noted a similar pigment in *Az. insigne* (unpublished observation).

Antisera active against *Az. chroococcum* have been prepared, and this work is being extended to include antisera to each of the species of *Azotobacter*. Attempts will be made to determine the chemical nature and position within the bacterial cell of some of these antigens, and a fuller report of the work will be published elsewhere.

I am indebted to Dr. J. G. P. Hutchison, of this Department, for advice concerning the drying and staining of gel diffusion plates.

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[1] Azo, K., and Yoshida, R., Proc. First Int. Congr. Soil Sci., 3, 150 (1928).
[2] Jensen, V., and Petersen, E. J., Roy. Vet. and Agric. Coll., Copenhagen, Den., Yearbook, 107 (1955).
[3] Petersen, E. J., Roy. Vet. and Agric. Coll., Copenhagen, Den., Yearbook, 70 (1959).
[4] Eisenstark, A., Ward, C. B., and Kyle, T. S., *J. Bact.*, 60, 525 (1950).
[5] Lawson, G. J., and Stacey, M., *J. Chem. Soc.*, 1925 (1954).
[6] Carr, L. B., and Varner, J. E., *Bact. Proc.*, 58 (1958).
[7] Jensen, V., *Bot. Tidsskr.*, 52, 143 (1955).
[8] Johnstone, D. B., *App. Microbiol.*, 5, 103 (1957).

Culture of *Desulphovibrio desulphuricans*

In a discussion about the method proposed by us[1] for the counting of sulphate-reducing bacteria in mixed population, Dr. J. R. Postgate pointed out in correspondence that, if one is handling a natural sample in which the population of sulphate-reducing bacteria out-numbers that of other micro-organisms, then the higher dilutions of such a sample are effectively pure cultures. Evaluation of our method with pure cultures of *D. desulphuricans* was obviously indicated. Dr. Postgate kindly furnished us with lyophilized pure cultures of the freshwater strain Hildenborough (N.C.I.B. 8303) and the salt-water strain El Agheila *Z* (N.C.I.B. 8380) of *D. desulphuricans*. Our experience in the culturing of both strains from the desiccates is summarized in Table 1; an iron nail in the medium clearly obviates difficulties in the revival of lyophilized cultures and, in addition, makes the exclusion of oxygen unnecessary. In a medium containing a nail, cysteine did not appear to be essential although it promoted earlier growth.

For comparing media for use in determining dilution counts of *D. desulphuricans*, decimal dilutions of a fresh culture in Starkey's medium (*LS*) were prepared in physiological saline, and 1 ml. of each dilution was transferred to each of five tubes of medium being tested. Incubation was at 30° C. for seven days. Some typical results are shown in Table 2. The results did not differ when cysteine hydrochloride (Merck) was included in *LS* + nail before sterilization or as Seitz-filtered solution just prior to inoculation. As autoclaving cysteine in presence of nail not infrequently resulted in slight darkening of the medium, it was found advantageous to add the flame-sterilized nail before inoculation. Neither Grossman and Postgate's[2] medium (*LSYC*) nor Starkey's medium containing a nail gave quantitative recoveries of viable bacteria. The inclusion of iron in the first medium or of an aerobic organism such as *Serratia marcescens* or *Aerobacter aerogenes* in the second permitted good growth. Addition of cysteine, yeast extract or peptone to *LS* + nail greatly improved the results. Such favourable effect was not apparent in the absence of nail. This suggests that in the presence of nail, cysteine acted chiefly as a nutrient (cf. Senez and Leroux-Gilleron[3]) rather than by poising the oxidation-reduction potential. The benefits given to aerobes do not seem to be simply due to oxidation-reduction potential changes as they gave occasionally poorer results than media containing cysteine and their effect was not apparent in the absence of nail. A question that requires elucidation is why the organisms in absence of iron nail found difficulty in initiating growth. It looks as if the nail provides an optimal surface for the initiation of growth, as the blackening

Table 1. INFLUENCE OF MEDIUM ON REVIVAL OF *D. desulphuricans* FROM LYOPHILIZED CULTURES*

Medium†	Anaerobic incubation‡	No. of tubes inoculated	No. of tubes showing growth§	Incubation period at 30° C. (days)
		Hildenborough strain		
LSYC	+	2	0	12
LSYC nail	+	3	3	2
LSYC nail	−	3	3	2
LSC nail	+	3	3	2
LSC nail	−	3	3	2
LS nail	+	4	4	3
LS nail	−	2	2	3
		El Agheila *Z* strain		
LSYC	+	2	0	9
LSYC nail	+	2	2	2
LSYC nail	−	2	2	2
LSC nail	+	2	2	2
LSC nail	−	2	2	2
LS nail	+	2	2	5
LS nail	−	2	2	8

* Inoculum *c.* 0·05 ml. per tube containing 5 ml. medium.

† *L*, sodium lactate, 3·5 gm./l.; *S*, sodium sulphate, 2 gm./l.; *Y*, yeast extract 'Difco', 1 gm./l.; *C*, cysteine hydrochloride, 1 gm./l. All media contained ammonium chloride, 1 gm.; dipotassium hydrogen phosphate, 0·5 gm.; calcium chloride, 0·1 gm.; Mohr's salt, 0·1 gm.; and tap water to 1,000 ml.; *p*H 7. For El Agheila *Z* strain 25 gm. sodium chloride per litre media were included.

‡ Provided by pyrogallol and sodium carbonate.

§ Indicated by blackening.

Table 2. GROWTH OF *D. desulphuricans* UNDER DIFFERENT CULTURAL CONDITIONS

Medium*	Anaerobic incubation	No. of positive tubes at a dilution of 10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}	10^{-6}	10^{-7}	10^{-8}	10^{-9}
		Hildenborough strain								
LSYC	+	5	0	1	0	1	0	0	0	0
LSYC nail	+	5	5	5	5	5	5	5	5	5
LSYC nail	−	5	5	5	5	5	5	5	4	5
LSY nail	−	5	5	5	5	5	5	5	3	1
LSP nail	−	5	5	5	5	5	5	5	5	1
LS nail	−	5	0	0	0	0	0	0	0	0
LS nail	+	5	0	2	2	0	0	0	0	0
LS nail *S. marcescens*	−	5	5	5	5	5	5	5	5	5
		El Agheila *Z* strain								
LSYC	+	0	0	0	0	0	0	0	0	0
LSYC nail	+	5	5	4	5	5	4	5	5	4
LSYC nail	−	5	5	3	5	4	5	5	5	2
LSY nail	−	5	5	5	5	5	5	5	2	0
LS nail	−	2	0	0	0	0	0	0	0	0
LS nail	+	3	0	0	0	0	0	0	0	0
LS nail *S. marcescens*	−	5	5	5	5	5	5	4	1	0

* See footnote to Table 1.

in the cultures invariably appeared first on the nail, and once growth is initiated the hydrogen sulphide produced helps the continuity of active multiplication because of its reducing properties. This interpretation finds support in the experiments on corrosion at Teddington in which growth was observed to start on the iron specimen (Postgate, J. R., personal communication).

Three media for dilution counts of *D. desulphuricans* were compared using soil containing a dense population of that species. The soil was prepared by placing 25 gm. of an air-dry, sieved clay loam in a tube, adding 6 ml. of a fresh *LS* culture of the 'Hildenborough' strain, and 6 ml. of sterile *LS* medium; the soil was then incubated anaerobically for 3 days at 30° C. Counts were then made using (*a*) *LS* + nail, (*b*) *LS* + nail, inoculated from one-day agar culture of *Serratia marcescens*, and (*c*) *LSYC* + nail. Results typical of those obtained in three trials appear in Table 3. They indicate that Starkey's medium plus nail is suitable for counting sulphate-reducing bacteria in natural materials containing large numbers of such organisms.

Table 3. DILUTION COUNTS OF DENSE POPULATIONS OF *D. desulphuricans* IN SOIL

Medium*	No. of positive tubes at a dilution of:† 10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}	10^{-6}	10^{-7}	10^{-8}	10^{-9}
LS+nail	5	5	5	5	5	5	3	1	0
LS+nail+ *S. marcescens*	5	5	5	5	5	5	3	2	0
LSYC+nail	5	5	5	5	5	3(2)	2(3)	0(5)	0

* See footnote to Table 1.
† Figures in parenthesis indicate false positive tubes.

We wish to thank Dr. T. Gibson of the School of Agriculture, Edinburgh, and Dr. J. R. Postgate of the Microbiological Research Establishment, Porton, for constructive criticism and valuable suggestions.

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[1] Abd-el-Malek, Y., and Rizk, S. G., *Nature*, 182, 538 (1958).

[2] Grossman, J. P., and Postgate, J. R., *Proc. Soc. App. Bact.*, 16, 1 (1953).

[3] Senez, J. C., and Leroux-Gilleron, J., *Bull. Soc. Chim. Biol.*, 36, 553 (1954).

GENETICS

Differential Transmission of Gametes in Wheat

ONE of the basic principles of Mendelian genetics is that union of different gametes takes place at random, and genetic ratios are based on this assumption. In a few species, including *Vigna sinensis* Endb.[1] and *Hordeum vulgare* L.[2], the possibility of differential transmission of alleles has been suggested. The work reported here produces evidence that differential gametic transmission operates in wheat and gives rise to irregular inheritance patterns.

Investigators in Australia and North America have found that the varieties Gabo, Yalta, Charter, Lee and Timstein carry the same resistance to wheat stem rust, *Puccinia graminis* Pers. var. *tritici* (Eriks. and E. Henn.). Reports differ, however, as to the mode of inheritance of resistance. Workers in North America[3], using Chinese Spring as the susceptible variety, found evidence for two linked, dominant, complementary factors governing resistance (*Sr* 11 and *Sr* 12). Monosomic analysis located the two genes on the X chromosome, and linkage values of 20–29·5 per cent were proposed. Australian workers[4,5], on the other hand, using crosses with Federation, concluded that a single factor is involved.

Most of the data for this study were obtained from F_2 and F_3 generations of crosses involving the varieties Gabo, Yalta, Charter, Eureka and Mentana. The latter two were used as stem rust susceptible parents. Mentana is resistant to strain 68 *Anz*-3 of wheat leaf rust, *Puccinia triticina* Erikss. Seedling tests with leaf rust were carried out and the plants classified as resistant, intermediate or susceptible. In their reaction to stem rust, plants were clearly either resistant or susceptible.

Failure to obtain susceptible segregates in the F_2 of intercrosses confirms that the varieties Gabo, Yalta and Charter have the same gene or genes for resistance to race 126 of stem rust. In crosses of these three varieties with Eureka a single, dominant gene seemed to segregate. Crosses with Mentana, however, gave results similar to those reported by North American workers. They did not fit a single-factor hypothesis, and in the cross Yalta × Mentana a ratio of approximately 1 : 1 was evident (Table 1).

Table 1

	Resistant	Susceptible	Ratio	Probability (3 : 1)
Gabo × Eureka	462	161	2·9 : 1	0·70–0·50
Charter × Eureka	89	31	2·9 : 1	0·90–0·80
Yalta × Eureka	186	61	3·0 : 1	0·95–0·90
Gabo × Mentana	404	169	2·4 : 1	0·02–0·01
Charter × Mentana	500	226	2·2 : 1	<0·001 (9 : 7)
Yalta × Mentana	467	465	1·0 : 1	<0·001

Similarly, in studies of leaf rust resistance, there was inconsistency in F_2 segregation of crosses involving Mentana and the varieties Eureka, Spica, Kenya 117*A*, Gabo, Charter and Yalta. In the latter cross only a few susceptible F_2 seedlings were found. It was also apparent that the stem rust resistance of Gabo, Yalta and Charter was closely linked with the leaf rust resistance of Mentana. As these resistances entered the crosses in the repulsion phase it was thought possible that the unusual F_2 ratios might be explained by examining F_3 lines (Table 2).

Table 2. REACTION OF F_3 LINES OF CROSSES GABO × MENTANA AND YALTA × MENTANA TO STRAINS 126 *Anz*-1 OF *P. gr. tritici* AND 68 *Anz*-3 OF *P. triticina*

		Reaction to *P. gr. tritici*					
		Resistant		Segregating		Susceptible	
Reaction to *P. triticina*	Resistant	—	—	—	—	62	107
	Segregating	—	1	121	93	3	2
	Susceptible	40	4	—	1	1	—

First columns, Gabo × Mentana; second columns, Yalta × Mentana.

The results shown in Table 2 cannot be explained on a Mendelian basis. In both crosses close linkage in the repulsion phase between stem rust and leaf rust resistance is evident, and the different segregation ratios in the two crosses indicate that differential transmission of gametes is involved.

A striking feature in the F_2 ratio of the cross Yalta × Mentana was the low proportion of homozygous stem rust resistant lines. This, like the previous F_2 ratio of 1 : 1, could be explained by partial failure of pollen carrying the dominant allele. In many segregating F_3 lines, however, there was a considerable excess of susceptible segregates, and a second test of 14 lines confirmed this behaviour. The actual numbers were 150 resistant to 294 susceptible (χ^2 for a 1 : 1 ratio $= 46\cdot703$; $P = <0\cdot001$). This indicates that differential transmission operates in the female as well as in the male gametes.

A backcross Mentana × (Yalta × Mentana) tested with stem rust gave 2 resistant and 11 susceptible plants. The two resistant plants, when selfed, gave progeny in a ratio of 2·2 resistant : 1 susceptible instead of the expected 1 : 1. This contradicts the hypothesis that the mode of inheritance of resistance of Yalta is controlled by two or three dominant, complementary factors, as this hypothesis requires that the ratio in backcrosses to Mentana remain constant at 1 : 1.

These studies support the suggestion that there is only one factor for Gabo-type resistance. It is also likely that there is only one major factor for leaf rust resistance in Mentana. The main basis for these assumptions is the relative infrequency of crossing-over in the repulsion phase.

That partial sterility is the cause of these results is improbable. F_1 plants of Mentana crosses show no marked infertility; and the high germinability of the F_2 seeds does not indicate differential zygotic viability.

Further work will be done to find the mechanism causing this departure from random Mendelian segregation. It would also be of interest to determine whether chromosomes other than the X chromosome are affected.

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[1] Smith, F. L., *Hilgardia*, **24**, 279 (1956).

[2] Luig, N. H., McWhirter, K. S., and Baker, E. P., *Proc. Linn. Soc., N.S.W.*, **83**, 340 (1958).

[3] Knott, D. R., and Anderson, R. G., *Canad. J. Agric. Sci.*, **36**, 174 (1956).

[4] Watson, I. A., and Stewart, D. M., *Agron. J.*, **48**, 514 (1956).

[5] Watson, I. A., and Waterhouse, W. L., *Proc. Linn. Soc. N.S.W.*, **74**, 113 (1949).

Correlation of Loss of Photoreversibility of Ultra-violet-induced Mutations with Deoxyribonucleic Acid Synthesis

PREVIOUS work suggests that nucleic acid precursors altered *in vivo* by ultra-violet light serve as metabolic intermediates for mutation induction in *Escherichia coli*[1]. Other studies indicate that in the absence of an adequate source of nitrogen, an immediate and rapid decline in frequency of mutations induced by ultra-violet light occurs[2,3]. Prevention of this decline depends on the availability of a complex supply of amino-acids immediately following radiation exposure. The potential mutation is modified during post-irradiation incubation so that it is no longer subject to reversal by those agents inhibitory to protein or ribonucleic acid synthesis which cause decline in mutation frequency. Progression of loss of mutation susceptibility to chloramphenicol reversal is closely correlated with the post-irradiation synthesis of ribonucleic acid. Furthermore, experimentation has shown that a relation exists between the amount of post-irradiation ribonucleic acid which has been synthesized at the time of addition of chloramphenicol, the rate of synthesis of deoxyribonucleic acid in the presence of chloramphenicol, and the induced mutation frequency response obtained at subsequent plating[4]. These results suggest that ribonucleic acid and protein syntheses are required for mutation induction and synthesis of deoxyribonucleic acid following exposure to ultra-violet light. It would appear that only those cells which have synthesized the ribonucleic acid and protein necessary for genetic replication prior to addition of chloramphenicol are able to reproduce their deoxyribonucleic acid and thereby acquire mutant characters.

Recent studies of mutation expression have yielded further information on the relation of mutation induction to synthesis of deoxyribonucleic acid[5]. Mutation expression closely follows the initial net synthesis of deoxyribonucleic acid in irradiated cultures. Since chloramphenicol treatment subsequent to synthesis of deoxyribonucleic acid blocks this expression, protein synthesis following synthesis of the acid must take place in order for the induced mutations to be phenotypically expressed. The experiments we wish to report here relate to the involvement of synthesis of deoxyribonucleic acid in the mutation-induction process. These studies demonstrate that loss of photoreversibility of ultra-violet-induced mutation is closely correlated with post-irradiation synthesis of deoxyribonucleic acid.

The mutations studied result in reversion to the prototrophic state in *E. coli* strain *WP2* (a tryptophan-

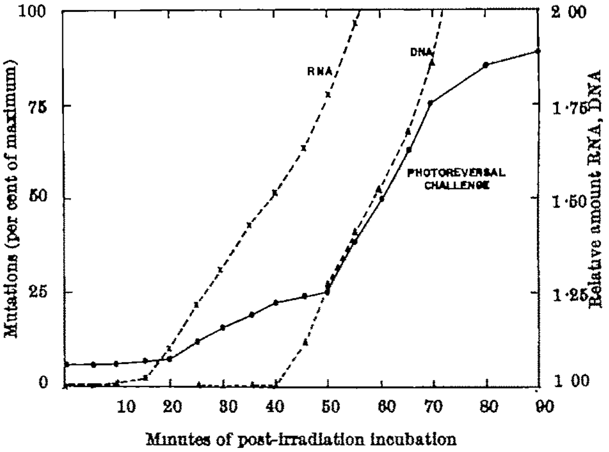

Fig. 1. The effect of challenge by photo-reversing light at increasing post-irradiation incubation intervals on the yield of ultra-violet-induced prototrophs from *Escherichia coli* strain *WP2*. Mutations are expressed as the percentage of those appearing in ultra-violet-irradiated control cultures which were incubated for an identical time interval and held for 1 hr. in an ice bath in the dark prior to plating. Photoreversal was carried out by subjecting ultra-violet-irradiated cultures to the light emitted by two 500-watt General Electric projection lamps. These were located 6 in. from the culture samples which were in small test-tubes immersed in an ice bath. The samples were exposed to the photo-reversing light for 1 hr., an exposure period which has been shown to produce maximum photoreversal. The incubation medium was minimal medium (ref. 1) plus casein hydrolysate (2 mgm./ml.) and DL-tryptophan (0·02 mgm./ml.). Ultra-violet exposure was 960 ergs/mm.^{2}. Induced mutation frequency response for unphotoreversed cells was approximately 8,000 prototrophs per 10^8 surviving organisms and did not vary significantly with incubation over a 90-min. period. Plating was on mutation assay agar previously described (see Doudney and Haas, ref. 3). Survival following this dose of ultra-violet radiation was approximately 50 per cent. The white light treatment resulted in photo-reactivation to the 70 per cent level of survival when applied without incubation. This effect was lost by 20 min. incubation. The cultures during incubation contained about 10^8 bacteria per ml. (as determined by colony formers observed with plating prior to radiation exposure)

requiring auxotroph isolated by Witkin). The procedures for culture growth, synchronization, radiation exposure, post-irradiation treatment, plating and mutation assay have been described, as have the procedures used for nucleic acid determination[1–3].

The loss of susceptibility of ultra-violet-induced mutations to photoreversal with post-irradiation incubation has been demonstrated with several systems[6–11] and a similar effect with the tryptophan reversion used here has been described[3]. Fig. 1 demonstrates that approximately 20 per cent of the potential mutations lose their capacity to be photoreversed prior to net measurable synthesis of deoxyribonucleic acid in the culture. However, the larger portion of potential mutations lose this capacity in correlation with synthesis of deoxyribonucleic acid. At the time that deoxyribonucleic acid has doubled (70 min.) approximately 80 per cent of the mutations are no longer susceptible to photoreversal.

Two processes may be involved in this loss of susceptibility to photoreversal. The initial process may represent some physical or chemical conversion of the radiation-modified factor responsible for the induction of the mutation to a non-photosensitive state prior to synthesis of deoxyribonucleic acid. However, the loss of susceptibility to photoreversal which is correlated with synthesis of deoxyribonucleic acid would seem to be due to actual synthesis of the mutated gene. These results therefore support the hypothesis that ultra-violet-induced mutation involves finally the synthesis of deoxyribonucleic acid following ultra-violet exposure[5].

These preliminary studies will be reported more fully elsewhere. This research was supported in part by research grant C-3323 from the United States National Institutes of Health and by U.S. Atomic Energy Commission Contract AT-(40–1)-2139.

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[1] Haas, F. L., and Doudney, C. O., *Proc. U.S. Nat. Acad. Sci*, 43, 871 (1957).

[2] Doudney, C. O., and Haas, F. L., *Proc. U.S. Nat. Acad. Sci.*, 44, 390 (1958).

[3] Doudney, C. O., and Haas, F. L., *Proc. U.S. Nat. Acad. Sci.*, 45, 709 (1959).

[4] Doudney, C. O., and Haas, F. L. (in preparation).

[5] Haas, F. L., and Doudney, C. O., *Proc. U.S. Nat. Acad. Sci.*, 45, 1620 (1959).

[6] Kelner, A., *J. Bacteriol.*, 58, 511 (1949).

[7] Newcombe, H. B., and Whitehead, H. A., *J. Bacteriol*, 61, 243 (1951).

[8] Novick, A., and Szilard, L., *Proc. U.S. Nat. Acad. Sci.*, 35, 591 (1949).

[9] Novick, A., Brookhaven Symp. Biol., 8, 201 (1955).

[10] Newcombe, H. B., Brookhaven Symp. Biol., 8, 88 (1955).

[11] Matney, T. S., Shankel, D. M., and Wyss, O., *J. Bacteriol*, 75, 180 (1958).

SOIL SCIENCE

Symbiotic Nitrogen Fixation in a Grazed Tropical Grass–Legume Pasture

The importance of legumes as symbiotic nitrogen-fixers in the pastures of temperate regions is generally recognized, and considerable data are available to show the contributions made to the soil nitrogen supply by these legumes. Very little information of this nature, however, is available in the case of legumes growing in tropical pastures. Tropical legumes are often considered to be inefficient nitrogen-fixers; in instances where this is so, it is probable that inoculation with the appropriate *Rhizobium* strain or the supplying of deficient nutrients, or both, will increase fixation efficiency. Results obtained from an experiment on the Faculty of Agriculture Farm, University College, Ibadan, indicate that under suitable conditions the legume in a pasture growing under equatorial conditions may contribute amounts of nitrogen to the soil similar to those added by legumes in temperate pastures.

A grazing experiment comparing the productivity of a pure grass stand (*Cynodon plectostachyum*) and a mixed pasture (*Cynodon plectostachyum–Centrosema pubescens*) was established in mid-1953 and terminated in mid-1958. The soil of the experimental area was a latosol, Ibadan loamy coarse sand. On inspection the roots of the *Centrosema* were found to be well nodulated. The colour of the grass in the mixed pasture was a much darker green than that of the pure grass plot, suggesting that there were significant soil nitrogen differences between these plots. In September 1958 the plots were sampled to a depth of 12 in. and total (Kjeldahl) nitrogen of the samples determined. These nitrogen contents are shown in Table 1; to express the values in lb. per acre it was assumed that an acre-6-in. of soil weighed 2 million lb.

Table 1. SOIL NITROGEN (KJELDAHL) UNDER FIVE-YEAR-OLD GRAZED PURE GRASS AND GRASS–LEGUME PASTURES AT UNIVERSITY COLLEGE, IBADAN, NIGERIA

Depth (in.)	Soil nitrogen under *Cynodon plectostachyum* and *Centrosema pubescens* (lb. per acre)	Soil nitrogen under *Cynodon plectostachyum* (lb. per acre)	Difference in soil nitrogen (lb. per acre)
0–2	770	510	260
2–4	460	370	90
4–8	770	610	160
8–12	600	550	50
0–12 (Total)	2,600	2,040	560*

Each value is the mean of three replicates.
* '*t*' test: $P = 0{\cdot}10$.

There were 560 lb. of nitrogen per acre-ft. more in the soil under grass–legume than in the soil under grass. This was a net gain over a five-year period and may reasonably be attributed to symbiotic nitrogen fixation by the *Centrosema*. On an average annual basis the *Centrosema* had fixed 112 lb. of nitrogen per acre-ft. Thus the average annual increment of soil nitrogen due to *Centrosema* in this tropical mixed pasture was of the same order of magnitude as those for temperate pasture legumes. For example, Russell[1] states that clovers and lucerne may add to the soil 150–200 lb. of nitrogen per acre per year.

It appears that, under suitable conditions, pasture legumes may play as important a part in the nitrogen economy of tropical soils as they do in the case of temperate soils.

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[1] Russell, E. J., "Soil Conditions and Plant Growth", 323 (Longmans, Green and Co., London, 1950).

FORTHCOMING EVENTS

(*Meetings marked with an asterisk * are open to the public*)

Monday, February 29

University of London (in the Physiology Theatre, University College, Gower Street, London, W.C.1), at 5 p.m.—Dr. Antoinette Pirie . "The Biochemistry of the Eye".* (Further lecture on March 7).

British Society for the History of Science (in the Lecture Theatre of the Science Museum, Exhibition Road, South Kensington, London, S.W.7), at 5.30 p.m.—Dr. W. Cameron Walker: "Foundations of Current Electricity and Electro-Chemistry".

Society of Chemical Industry, Surface Activity Group (at 14 Belgrave Square, London, S.W.1), at 5 30 p.m.—Dr. E. G. Cockbain: "The Colloidal Stability of Natural Rubber Latex".

Royal Geographical Society (at 1 Kensington Gore, London, S.W.7), at 8.30 p.m.—Mr. A. J. Cain: "The Oxford University Expedition to British Guiana".

Tuesday, March 1

University of London (in the Anatomy Theatre, University College, Gower Street, London, W.C.1), at 1.15 p.m.—Dr. A. J. Arkell: "An Archaeologist Visits the Belgian Congo".*

Institution of Electrical Engineers, Measurement and Control Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Scientific Papers.

Royal Society of Medicine, Pathology Section (at 1 Wimpole Street, London, W.1), at 5.30 p.m.—Prof. L. S. Penrose, F.R.S. . "Human Genetics and Disease"; Dr. H. Lehmann: "Familial Deficiency of Pseudocholinesterase"; Dr. A. Holzel · "Galactosæmia—a Hereditary Disease".

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Dr. A. Isaacs . "The Anti-Viral Action of Interferon". (Last of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation.)*

Institution of Mechanical Engineers, Automobile Division (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Mr. Wheeler G. Lovell: "The Chemistry of Future High-Compression Engines" (James Clayton Lecture).

University of London (in the Beveridge Hall, Senate House. London, W.C.1), at 6 p.m.—Sir Douglas Logan: Birkbeck College Haldane Memorial Lecture.

Plastics Institute (at the Wellcome Building, 183–193 Euston Road, London, N.W.1), at 6.30 p m.—Dr. N. Grassie: "Recent Investigations into Polymer Degradation".

Society of Chemical Industry, Plastics and Polymer Group (at 14 Belgrave Square, London, S.W.1), at 6.30 p.m.—Dr. A. Keller: "Some New Aspects of the Crystallization of High Polymers".

Wednesday, March 2

Royal Society of Arts (at John Adam Street, Adelphi, London, W.C.2), at 2.30 p.m.—Mr. F. I. G. Rawlins: "Scientific Methods in the Care of Works of Art".

Institute of Petroleum (at 61 New Cavendish Street, London, W.1), at 5.30 p.m.—Mr. Michel Estavoyer: "Corrosion Problems in the Lacq Field".

Royal Meteorological Society (at 49 Cromwell Road, London, S.W.7), at 5.30 p.m.—Dr. H. L. Penman: "Weather, Soil and Plant Factors in Hydrology".

University of London (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. G. Schramm (Tubingen): "Mutagenesis in Viruses".*

Institution of Mechanical Engineers, Nuclear Panel (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Discussion on "The Role of Formal Education and Training |in Nuclear Power Developments".

Thursday, March 3

Royal Society (at Burlington House, Piccadilly, London, W.1), at 10.30 a.m.—Discussion on "Problems of Stellar Evolution", opened by Prof. F. Hoyle, F.R.S.

Institute of Refrigeration (at the Institute of Marine Engineers, The Memorial Building, 76 Mark Lane, London, E.C.3), at 5.30 p.m.—Dr. E. Baumgartner: "European Refrigerated Transport by Railway".

Institution of Electrical Engineers (joint meeting with the Institutions of Civil and of Mechanical Engineers, at Savoy Place, London, W.C.2), at 5.30 p.m.—Sir Hugh Beaver, K.B.E. . "The Engineer and Civilization" (Sixth Graham Clark Lecture).

British Psychological Society, Education Section (in the University of London Institute of Education, Malet Street, London, W.C.1), at 6.15 p.m —Annual General Meeting. Prof. Allen L. Edwards (U.S.A.): "The Effects of Social Desirability upon Personality Measurement".

Friday, March 4

Zoological Society of London (at Regent's Park, London, N.W.1), at 10.15 a.m.—Symposium on "Sensory Specialization in Response to Environmental Demands".

Linnean Society of London (joint meeting with the Systematics Association, at Burlington House, Piccadilly, London, W.1), at 10.45 a.m.—Symposium on "The Subspecies". Speakers include Prof. Valentine, Dr. Hawkes, Dr. Warburg, Mr. Ross, Dr. Galbraith, Dr. Clay, Mr. Stroyan and Dr. de Wolf.

Royal Astronomical Society (joint meeting with the Royal Meteorological Society, at the Royal Society of Arts, John Adam Street, London, W.C.2), at 2.30 p.m.—Geophysical Discussion on "The British Rocket Programme". Chairman: Prof. Sir H. S. W. Massey, F.R.S.).

Institution of Electrical Engineers, Medical Electronics Discussion Group (at Savoy Place, London, W.C.2), at 6 p.m.—Discussion on "Direct Writing Oscillographs" opened by Mr. A. Smale and Mr. S. N. Pocock.

Society of Dyers and Colourists (at the Northampton College of Technology, St. John Street, London, E.C.1), at 6 p.m.—Mr. J. H. Sharphouse: "Recent Developments in Leather Dyeing".

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Sir Charles Darwin, K.B.E., F.R.S.: "Some Episodes in Darwin's Voyage on 'The Beagle'".

Saturday, March 5

British Interplanetary Society (in the Lancaster Room, Caxton Hall, Westminster, London, S.W.1), at 2.30 p.m.—Symposium on "The Exploration of the Moon".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Assistant Lecturer (preferably with research interest in polymer chemistry, carbonyls or high-pressure reactions) in Organic Chemistry—The Registrar, The Manchester College of Science and Technology, Manchester 1 (March 5).

Lecturer in the Philosophy of Science—The Secretary, The University, 38 North Bailey, Durham (March 7).

Lecturer in Physical Chemistry; and an Assistant Lecturer in Zoology—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1 (March 8).

Senior Scientific Officer, Physical Chemist (with a first- or second-class honours degree or equivalent) at Warren Spring Laboratory, D.S.I.R., Stevenage, Herts, for work on electrochemistry of surface and colloidal reactions, forming part of a programme of research on fundamentals of froth flotation and mineral processing—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. F.79/0A (March 11).

Assistant Lecturers (2) (interested in plant biochemistry, experimental work with marine algae or genetics) in Botany; a Lecturer or Assistant Lecturer in Inorganic Chemistry; an Assistant Lecturer in Physical Chemistry; and an Assistant Lecturer in Organic Chemistry; an Assistant Lecturer in Geography; an Assistant Lecturer in Pure Mathematics and an Assistant Lecturer in Applied Mathematics; a Senior Lecturer in Physics; an Assistant Lecturer in Vertebrate Embryology and an Assistant Lecturer in Invertebrate Zoology—The Registrar, The University, Hull (March 12).

Curator of the Gulbenkian Museum of Oriental Art and Archæology—The Secretary, The University, 38 North Bailey, Durham (March 12).

Hospital Physicist, Basic or Senior grade (preferably with a knowledge of radiotherapy)—The House Governor, King's College Hospital, Denmark Hill, London, S.E.5 (March 12).

Lecturer (preferably with industrial experience or an interest in applied physical chemistry) in Physical Chemistry in the Department of Applied Chemistry—The Secretary, Northampton College of Advanced Technology, St. John Street, London, E.C.1 (March 12).

Lecturer, Terminable Lecturers, and an Assistant in the Department of Mathematics—The Secretary, The University, Aberdeen (March 12).

Assistant Lecturer in Physics—The Secretary and Registrar, University College of North Wales, Bangor, North Wales (March 15).

Lecturer (with a special interest in one or more of the following fields: soils, conservation, or land use) in Geography—The Secretary, University College, Gower Street, London, W.C.1 (March 15).

Lecturer (preferably with a special interest in semi-conductors and their applications) in Applied Physics—The Secretary, The University, 38 North Bailey, Durham (March 18).

Assistant Lecturer in Endocrine Biochemistry, to work in the University Endocrine Unit at the Liverpool Radium Institute—The Registrar, The University, Liverpool, quoting Ref. CV/T (March 19).

Lecturer (with some experience in microscopic anatomy or electron microscopy) in Anatomy—Joint Clerk to the University Court, Queen's College (University of St. Andrews), Dundee (March 19).

Lecturer or Assistant Lecturer (with special qualifications in palæontology or stratigraphy) in Geology—The Registrar, The University, Sheffield (March 19).

Lecturer in Psychology—The Secretary, The Queen's University, Belfast (March 21).

Lecturer in Physics at the University of Cape Town, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa, March 22).

Chair of Metallurgy—The Registrar, University College of South Wales and Monmouthshire, Cathays Park, Cardiff (March 26).

Lecturer in Botany—The Secretary, The University, 38 North Bailey, Durham (March 26).

Lecturer (with research experience in physical oceanography or in an appropriate branch of physics or mathematics) in Physical Oceanography—The Secretary and Registrar, University College of North Wales, Bangor, North Wales (March 29).

RESEARCH FELLOW (with research experience in biochemistry or chemistry and interest in the physico-chemical aspects of biochemistry) IN THE DEPARTMENT OF PHYSICAL BIOCHEMISTRY, John Curtin School of Medical Research, Australian National University—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (March 30).

CHAIR OF MEDICINE tenable at Charing Cross Hospital Medical School—The Academic Registrar, University of London, Senate House, London, W.C.1 (March 31).

LECTURER or SENIOR LECTURER (with research experience in statistics) IN MATHEMATICS—The Secretary, The University, 36 North Bailey, Durham (March 31).

LECTURER IN PSYCHOLOGY at the University of Adelaide, Australia —The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia, March 31).

PRINCIPAL (with high academic qualifications and appropriate educational and administrative experience) OF THE NORTHERN POLYTECHNIC—R. H. Currell, F.C.A., Northern Polytechnic, Holloway, London, N.7 (March 31).

SENIOR LECTURER IN THEORETICAL PHYSICS at the University of Cape Town, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa, March 31).

READER IN PHYSIOLOGY at King's College, Strand, London, W.C.2 —The Academic Registrar, University of London, Senate House, London, W.C.1 (April 1).

READER IN MATHEMATICS at King's College, Strand, London, W.C.2—The Academic Registrar, University of London, Senate House, London, W.C.1 (April 4).

ELECTRON MICROSCOPIST IN THE DEPARTMENT OF PHYSICAL BIOCHEMISTRY, John Curtin School of Medical Research, Australian National University—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, April 18).

HEAD (with a good honours degree, preferably in physics or chemistry, experience of teaching and examinations at G.C.E. advanced level and also at degree level) of the Department of Science, Zaria Branch, Nigerian College of Arts, Science and Technology—The Council for Overseas Colleges, 12 Lincoln's Inn Fields, London, W.C.2 (April 29).

LECTURER IN PHYSIOLOGY (HISTOLOGY)—The Secretary of University Court, The University, Glasgow (April 30).

ACADEMIC REGISTRAR (graduate in science or arts, with teaching, industrial and administrative experience, and preferably knowledge of the work of a major technical college)—The Principal, College of Science and Technology, Ashley Down, Bristol 7.

ASSISTANT CLINICAL BIOCHEMIST (basic grade)—The Group Secretary, Booth Hall Children's Hospital, Blackley, Manchester 9.

BIOCHEMIST TO WORK IN THE ENDOCRINE LABORATORY, Obstetric Unit, mainly on steroids—The Secretary, University College Hospital Medical School, University Street, London, W.C.1.

CHEMIST (with a good honours degree in chemistry or equivalent professional qualification, and preferably specialization or experience in public analyst work) IN THE GOVERNMENT CHEMIST'S DEPARTMENT, Tanganyika, for general analytical work in the fields of toxicology, food and drugs, and water, and the supervision of assistants—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.117/8/037.

GRADUATE (with a knowledge of animal nutrition and/or agricultural chemistry) AT THE ANIMAL HEALTH TRUST FARM LIVESTOCK RESEARCH CENTRE, Stock, Essex, to assist in biochemical investigations—The General Secretary, Animal Health Trust, 14 Ashley Place, London, S.W.1.

LECTURER (preferably with university teaching experience and an active research interest) IN BOTANY—The Registrar, Queen Mary College (University of London), Mile End Road, London, E 1.

LECTURER (qualified to teach botany to B.Sc. and Diploma in Technology level, and preferably special qualifications in plant physiology or plant biochemistry) IN BOTANY—The Registrar, Bradford Institute of Technology, Bradford 7.

STATISTICIAN (with an honours degree in statistics or in mathematics with a diploma in statistics, and interested in the field of physiology) AT THE MEDICAL RESEARCH COUNCIL CLIMATE AND WORKING EFFICIENCY UNIT, Oxford—The Secretary, M.R.C. Unit, Department of Human Anatomy, South Parks Road, Oxford.

REPORTS and other PUBLICATIONS

(*not included in the monthly Books Supplement*)

Great Britain and Ireland

Department of Scientific and Industrial Research. Road Research 1958: The Report of the Road Research Board with the Report of the Director of Road Research. Pp. iv+83+4 plates. (London: H.M. Stationery Office, 1959.) 5*s* 6*d*. net. [2212

Mental Health Research Fund. Supplement Report and Balance Sheet with Income and Expenditure Account for the period from 1st October 1958 to 31st March 1959. Pp. 14. (London: Mental Health Research Fund, 1959.) [2212

National Research Development Corporation. Report and Statement of Accounts for the year 1st July, 1958 to 30th June, 1959. Pp. ii+22. (London. H.M. Stationery Office, 1959.) 1*s*. 3*d*. net. [2212

Final Act of the Conference on Antarctica together with the Antarctic Treaty. Pp. 12. (Cmnd. 913.) (London: H.M. Stationery Office, 1959.) 9*d*. net. [2212

The North Staffordshire Field Club. Transactions and Annual Report, 1957–58. (Vol. XCII.) Edited by H. V. Thompson. Pp. 71+1 plate. (Stoke-on-Trent: North Staffordshire Field Club, 1959.) 15*s*. [2212

Bulletins of Marine Ecology (formerly Hull Bulletins of Marine Ecology). Vol. 4 (November, 1959). Title Page; Contents; List of Personnel; and Explanation by Dr. G. T. D. Henderson. Pp. i–xiii. (Edinburgh: Scottish Marine Biological Association, 1959.) 5*s*. [2212

Other Countries

United States Department of the Interior: Geological Survey. Bulletin 1056–B: Index to the Geologic Names of North America. By D. Wilson, G. C. Keroher and Blanche E. Hansen. Pp. iv+407–622. 60 cents. Bulletin 1058–F: Geology and Coal Resources of the Homer District, Kenai Coal Field, Alaska. By Farrell F. Barnes and Edward H. Cobb. Pp. iv+217–260+plates 17–28. Bulletin 1072–F: Mineral Occurrences of New York State with selected references of Each Locality. By Elaine M. Luedke, Chester T. Wrucke and John A. Graham. Pp. iii+385–444+plate 9. Bulletin 1077: Geology of the Lake Mary Quadrangle, Iron County, Michigan. By Richard W. Bayley. Pp. v+112+7 plates. Bulletin 1081–B: Stratigraphy of the Inyan Kara Group in the Black Hills. By Karl M. Waagé. Pp. iii+11–90+plate 2. 40 cents. Bulletin 1092: Stratigraphy of Middle Tertiary Rocks in Part of West-Central Florida. By Wilfred J. Carr and Douglas C. Alverson. Pp. iv+111+3 plates. 1.25 dollars. Bulletin 1097–B: Lead-Alpha Age Determinations of Accessory Minerals of Igneous Rocks (1953–1957). By Howard W. Jaffe, David Gottfries, Claude L. Waring and Helen W. Worthing. Pp. iii+65–148. 30 cents. Bulletin 1107–A: Bibliography of U.S. Geological Survey Reports on Uranium and Thorium, 1942 through May 1958. By Paul E. Soister and Dora R. Conklin. Pp. v+167. 50 cents. Bulletin 1094: Geology of Possible Petroleum Provinces in Alaska. By Don J. Miller, Thomas G. Payne and George Gray. Pp. v+131+3 plates. 2 dollars. (Washington, D.C.: Government Printing Office, 1959.) [3012

United States Department of the Interior: Geological Survey. Water-Supply Paper 1309: Compilation of Records of Surface Waters of the United States through September 1950. Part 6–A: Missouri River Basin above Sioux City, Iowa. Prepared under the direction of J. V. B. Wells. Pp. v+672+1 plate. Water-Supply Paper 1320–D: Floods of January 1953 in Western Oregon and Northwestern California. By S. E. Rantz. Pp. v+321–339. Water-Supply Paper 1511: Surface Water Supply of the United States 1957. Part 7: Lower Mississippi River Basin. Prepared under the direction of J. V. B. Wells. Pp. xi+527. 1.75 dollars. Water-Supply Paper 1515: Surface Water Supply of the United States 1957. Part 2: Pacific Slope Basins in California. Prepared under the direction of J. V. B. Wells. Pp. xiii+634. 2 dollars. Water-Supply Paper 1542–A: Flow-Duration Curves. By James K. Searcy. (Manual of Hydrology: Part 2—Low-Flow Techniques) Pp. iv+33. 20 cents. Water-Supply Paper 1459–A: Survey of Ferrous-Ferric Chemical Equilibria and Redox Potentials. By J. D. Hem and W. H. Cropper. Pp. iii+31. 15 cents. Water-Supply Paper 1483: Geology and Ground-Water Resources of the Upper Lodgepole Creek Drainage Basin, Wyoming. By L. J. Bjorklund. Pp. iv+40+2 plates. (Washington, D.C.: Government Printing Office, 1959.) [3012

United States Department of the Interior: Geological Survey. Professional Paper 317–A: Geology of the San Juan Metropolitan Area, Puerto Rico. By Clifford A. Kaye. Pp. ix+48+9 plates. Professional Paper 317–B: Shoreline Features and Quaternary Shoreline Changes, Puerto Rico. By Clifford A. Kaye. Pp. v+49–140+plates 1, 10 and 11. 1.50 dollars. Professional Paper 320: Geochemistry and Mineralogy of the Colorado Plateau Uranium Ores. By Robert M. Garrels and Esper S. Larsen. Pp. v+236+8 plates. 2 dollars. (Washington, D.C.: Government Printing Office, 1959.) [3012

Tea Research Institute of East Africa. Annual Report for the year 1958. Pp. 49+1 plate. (Nairobi: Tea Research Institute of East Africa, 1959.) [3012

Publicaciones del Instituto Antituberculoso "Francisco Moragas" de la Caja de Pensiones para la Vejez y de Ahorros. Volumen XIII: La Extrana Biologia de las Plaquetas Sanguineas (Cultivos *in vitro*). By Dr. C. Xalabarder. Pp. 94 (64 figures). (Barcelona: Instituto Antituberculoso "Francisco Moragas", 1959.) [3012

Metropolitan Life Insurance Company. Statistical Bulletin, Vol. 40 (October 1, 1959): Rapid Suburban Population Growth; Hearing Impairment is Common; Mortality and Social Class. Pp. 12. (New York. Metropolitan Life Insurance Company, 1959.) [3012

Western Australia. Report of the Geological Survey Branch for the year 1957. Pp. 7. (Extract from the Report of the Department of Mines.) (Perth: Government Printer, 1959.) [3012

Comptes Rendus des Travaux du Laboratoire Carlsberg. Vol. 31, No. 14: Delayed Division in Tetrahymena Pyriformis Induced by Temperature Changes. By H. Thormar. Pp. 207–226. 2 Kr. 60 øre. Vol. 31, No. 15: Appearance of New N-terminal Residues Upon Treatment of Human and Bovine Serum Albumin with N-bromosuccinimide. By T. Peters, Jr. Pp. 227–235. 1 kr. 70 øre. Vol. 31, No. 16: Deuterium Exchange Studies of Sperm Whale Metmyoglobin. Pp. 235–249. 2 kr. 25 øre. Vol. 31, No. 17. Some Observations on Pentose Utilization by *Candida tropicalis*. By Helena Karczewska. Pp. 251–258. 1 kr. 50 øre. (Copenhague: Danish Science Press, Ltd. 1959.) [3012

Bureau des Longitudes. Annuaire pour l'An 1960, publié par le, Bureau des Longitudes. Pp. viii+692+4 planches. (Paris: Gauthier-Villars, 1959.) [3012

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THE WORTH OF THE INDIVIDUAL IN MODERN SOCIETY

THE annual reports of successive presidents of the Carnegie Institution of Washington have earned a high reputation for the prescience and imagination with which they survey the varied aspects of the inter-relations of science and society and the contribution which the independent research institution can make. Nor are such problems surveyed from a narrow or nationalistic point of view, and it is not surprising that these reports are read with as much appreciation by British as by American readers, or that there is frequently to be discerned a striking parallelism between the trend of thought in the president's report and that disclosed in such current addresses of British men of science as the anniversary address to the Royal Society, a Reith Lecture, or a university lecture like the Fawley Foundation Lecture at the University of Southampton. Dr. Caryl P. Haskins's current report* is no exception, and its opening section, in which he considers the challenge which is offered to men and nations by times of severe stress, of profound change and radical adjustment, such as those in which we live, may profitably be considered alongside some passages in Lord Adrian's recent Fawley Lecture, "The Risks of Progress".

Dr. Haskins begins by pointing out that hazard is not the only, or even the dominant, quality of such times, which offer far more than mere tests of ingenuity in living to be met by essentially conventional, if strenuous, feats of adaptation and accommodation. If met with boldness, with balance and dedication, and above all with independence of mind and spirit, such times may also be times of unmatched opportunities for new orders of growth and self-realization, the ultimate benefits of which could far outweigh the short-term perils that pre-occupy and shadow our generation. Earlier generations living in times of comparable stress and change recognized the uncertainty and danger of their lives, but they realized also the enormous challenge of their days. To-day a new eagerness and audacity can be sensed, and a flavour of the immense significance of individual effort and achievement has come forward to our day. Each period is coloured by a certain dynamic fluidity —a special ability, possibly a special eagerness, to embrace the condition of change for its own sake, regardless of its specific content—and all seem to have attached a higher value to the dynamic process of living in their society than to the material consequences. New ideas were valued and exciting, and their communication was as important as their genesis. Exploration, or the search for innovation in one or another realm of experience, was specially valued.

No patterns, of course, as Dr. Haskins reminds us, are ever precisely repeatable, and although it is not surprising that we should find in our own age certain parallels with other ages which have been characterized, in the perspective of history, as creative or successful, in certain ways, he rightly adds, our challenge differs from that confronting earlier periods so greatly in degree as perhaps to be of another kind. In no earlier society can the challenge to acuity of comprehension, to specialized skills, to generalized insight, to the fullest development of individual capability in every field, have been so imperative, nor can the demand and the opportunity for individual development have approached those of our own day. In our age surely the stimulus, the demand and the opportunities for innovation, for individual creativeness, have reached unprecedented heights.

Dr. Haskins next directs attention to a qualitative difference in the practical technical demands of our times. Earlier dynamic ages have probably put first in their technical strategy the search for sources of physical power. Now the very success of that quest suggests that in future other aspects of our conquest of Nature, concerned more particularly with understanding and control, will assume a more conspicuous place in our technology, as is already happening. In the future, they must emphasize further and give a keener incentive to a broader, perhaps subtler and deeper comprehension of Nature as a practical requirement of living.

Those observations point to the vital importance of education in the world to-day and to the soundness of the philosophy displayed in the first part of the recent Crowther Report. Dr. Haskins goes on to bring this out even more clearly when he shows that for nations, as for men, times of unusual stress and challenge are a supreme test, not alone of effectiveness in action, but also and primarily of inner cohesion and verve, of inner wholeness. The severest practical challenge can be met successfully by a people only in its own characteristic fashion, consonant with its own most deeply held values. Unless these are adequate and are undegraded, the whole strength and balance of a nation will be inadequate.

We are not, Dr. Haskins reminds us, immune from the dangers which swiftly eroded some of the great ages of the past. We are faced by practical technical challenges of a national and an international character in which the search for knowledge and understanding of Nature for its own sake is often linked with material achievements indispensable to our prosperity, or even essential for survival. In such a situation it is easy to mistake the end of the road for the road itself, and it is imperative accordingly that we should be clear about our deepest motivations, that we meet these challenges in our own way and by courses that will reinforce our own ethic. Whether we are individually directly concerned or not with scientific matters, we must comprehend the values that underlie the search for scientific verities and appreciate how vividly they mirror our more generalized ethic.

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* The Report of the President of the Carnegie Institution of Washington, 1958–1959. Pp. 54. (Reprinted from *Carnegie Institution of Washington Year Book* 58, for the year 1958–1959.) (Washington, D.C.: Carnegie Institution of Washington, 1959.)

The three points of anchorage which Dr. Haskins picks out from the philosophy of American society for special emphasis are no less part of the philosophy and tradition of Britain. First, there is that primary dedication to the individual and to his creativeness, that over-riding commitment which reveals the national stature primarily as a function of the welfare and perfection of realization of the worth and stature of the individual. This, Dr. Haskins writes, has been America's particular national pride to cultivate and foster, and on it depend the fundamental orientation and integrity of her society.

The second and third, like the first, may equally be regarded as part of America's heritage from Britain. The basically religious culture of America is closely linked with the first element, and though the dedication to the individual may be identified too narrowly with the Calvinist tradition from New England, it is undoubtedly largely derived from roots that are not materialistic. The third well-spring is a deep respect for, and concern with, both learning and innovation, and especially the exploration of Nature. The American people have traditionally loved the challenge of technical change; but equally the search for natural knowledge purely for its own sake is deeply indigenous in American culture.

Dr. Haskins recognizes, of course that in doing this science does no more than any other creative intellectual activity; but nevertheless, in its dedication of individual effort, its primary seeking after universally communicable knowledge of the natural world, in the discipline of its economy and its parsimony, and in its sense of style, scientific research must be reckoned to-day as one of the essential bulwarks of the deepest national values of the United States. That is largely true of other Western democracies, and in those democracies also material success offers a strong and continuing threat to such a non-materialistic ethic. It is not only in the United States that society needs a more explicit image of the real nature of its deepest motives—an image to which science may have an important part to contribute.

At this point Dr. Haskins touches on the problem of the two cultures as outlined by Sir Charles Snow. Whether or not this gulf arises from the confusion of application with creation—of regarding the scientific process as primarily an ever-expanding fount of technical product—the most serious danger is not in the field of education or communication, but rather that the divergence of experience may set and harden a real and fundamental dichotomy of attitudes and values between the two worlds, and encourage an unbridgeable hostility between them. The threat of such a fundamental division between the scientific and non-scientific world arises primarily from the divergence or misunderstanding of values rather than from unfamiliarity of subject-matter. Such a sense of division is inherently dangerous, and Dr. Haskins emphasizes some of the more serious hazards.

First, noting the consistent exponential growth of fundamental research since the time of Newton, doubling in volume approximately every ten or fifteen years, and at an appreciably greater rate than many other activities, he suggests that such a range and sweep of scientific effort must involve not merely the re-making of our culture materially but also the depletion of other creative activities in art, literature and humanism. This, of course, is one of the aspects of university expansion, particularly with regard to scientists and technologists, which already causes much concern in academic circles and elsewhere. If the gulf between the realms of science and other departments of human activity is indeed unbridgeable, involving irreconcilable differences of value and motive, the position is serious, for it implies that science must inevitably be permanently competitive with other areas of human effort.

While this could reduce the multivariant range of skills and interests which society needs to-day—and must indeed seek to encourage and expand—Dr. Haskins can point to evidence on the other side. Good minds in science, for example, tend also to be naturally good minds in other fields, and often to seek spontaneously the common denominators among them. Again, some of the most impressive scientific disciplines of to-day can be traced back historically in an almost unbroken evolution of ideas to representations of beauty and order in Nature on the part of eighteenth-century observers, in a pattern which to-day we might characterize as more artistic than scientific.

Even more significantly, perhaps, Dr. Haskins refers to the way in which, one by one, over the years, the barriers of attitude, of ways of thinking and modes of attack which, since the time of Newton, have traditionally segregated the concerns of science, put them in contrast with, and set them apart as narrower and somewhat different from, those of the rest of the world, are, in the larger philosophical sense, being penetrated. Post-Newtonian scientists knew, or soon discovered, that they must restrict their framework of scientific investigation to a limited segment of the world if their methods were to be applied successfully, and they ultimately paid with a highly disciplined parochialism for added depth and penetration; the notion of an unbridgeable dichotomy between scientific and extra-scientific orientation and concerns may be in part a consequence. A dramatic widening of the areas of scientific concern, with a progressive lowering of old barriers, however, is one of the major characteristics of our age, and it is conceivable that our notions about the separateness of the concerns and the domain of science from the rest of the world may become further blurred and ultimately fade.

When and if that occurs, it will come about, not by one view of the world replacing another, but rather by a convergence on common interests and a common kingdom. Such a development may prove to be the most powerful of all levellers of barriers, the most effective of any bridges spanning the gulf between the scientific and non-scientific points of view in the years to come. Unlike Lord Adrian, Dr. Haskins is unconcerned by the way in which in our time academic science and its relation to technology and industry have been violently forced out of the pattern which Huxley prescribed for science in the

Victorian period. The risks of progress at which he looks are intellectual rather than physical, and he believes that our concern that the exponential growth of scientific effort may disastrously pre-empt our human resources of mind and spirit may prove to be largely irrelevant.

Uncommitted scientific research, however, has a broader and deeper task than the winning of new knowledge, and apart from its contribution to essential human values, within its framework of opportunities, its breadth of horizon, its range of concepts and of problems, it has an important general obligation to discharge. In a vast technological society such as ours, bigness rather than success threatens the sanctity and maintained sense of worth of the individual. Survival depends on skilfully and successfully marshalling the work of myriads of individuals into the achievements of organized groups, and it is far too easy to forget that the very essence of creativeness must lie with the individual as contrasted with the group; the greatest strength of the group may be, at another time and in another place, to develop the full consequences of the vision of the individual.

That, Dr. Haskins insists, in conclusion, is what the research environment must stand for. To provide for the probity and the freedom of choice and action, to furnish the dynamic facilities for broad contacts and wide communication; and simultaneously, the shielding and opportunity for isolation that are so important to innovation and so precious at its most sensitive and valuable point; to foster flexibility and growth in the disciplines with which it is concerned—these are important and essential tasks. Yet more widely and profoundly significant, however, is its duty of manifesting its spirit as a society devoted to and sustained by the earned dignity of the individuals composing it. This is indeed the image of our larger society, which, as that society must be constantly reminded, alone can give it meaning, autonomy and authority. However successfully such a society may avert the physical risks on which Lord Adrian touched, it will not long endure unless it looks within and ensures that spiritual renewal and maintenance of values for which Dr. Haskins pleads so earnestly and well.

ZOOLOGICAL BORDERLANDS

On the Track of Unknown Animals

By Dr. Bernard Heuvelmans. Translated from the French by Richard Garnett. Pp. 558+52 photographs. (London: Rupert Hart-Davis, Ltd., 1959.) 35*s*. net.

BOOKS, like fashions, seem to follow cycles. The past decade has seen the appearance of several works devoted largely to speculation on the identity of mysterious unknown animals, descriptions of 'living fossils', and accounts of the more striking of the zoological discoveries of the present and past century. Possibly the sensational discovery of *Latimeria* in 1938, and the interest aroused by reports of unknown ape-like creatures in the Himalayas, have helped to popularize such topical books. In his preface, Dr. Heuvelmans has acknowledged his indebtedness to earlier writers in the same field; but it must be acknowledged that his is the most comprehensive, the best written, and certainly the most challenging work of its kind. The amount of research, as indicated by the extensive bibliography, must have been enormous, ranging from early classical writers to modern travellers and naturalists. Whether Dr. Heuvelmans is discussing the legendary Nittaewo (the lost people of Ceylon), the abominable snowman himself, or the supposed great aquatic reptile of Central Africa, his net is spread far and wide to bring in all available accounts and evidence. His ingenuity in linking legends, native stories or hunters' tales to produce a composite picture of some animal not yet recognized by science is praiseworthy, and certainly produces a great deal of engrossing reading matter.

But enthusiasm for the subject, allied to a fertile imagination, has resulted in a certain one-sidedness of approach, and many of his suggestions and conclusions, ingenious as they may be, lead to a feeling that the reverse of the medal has not been shown, or has been too briefly mentioned. It is the author's expressed conviction that some at least of the larger animals believed to have been long extinct are still surviving, and that there are also still unidentified large creatures, known only from native reports, or other hearsay evidence, concealed in the less well explored regions of the Earth. The many important zoological discoveries of the past hundred years or more are, in his view, only an indication of many more yet to come, rather than the natural consequence of the rapid opening up of so many of the wilder places of the Earth during that period.

It is in the development and illustration of this theme that the author allows his antipathy to the professional zoologist to colour all his views and warp his judgment. It appears that the professional zoologist, almost without exception, has resolutely turned a deaf ear to all reports of unknown animals, and has stubbornly refused to consider the possible existence of any creature the bones or skin of which are not on the museum table. In defence of the maligned professional, it should not be necessary to point out that it is only in relation to the established and known that the unestablished and unknown can be considered, and that the essential reference works and catalogues listing the multitudinous life around us are largely the work of the professional systematists. The accusation that zoologists are more concerned with adding names to the literature on insufficient grounds comes ill from a writer who, in this book, proposes three new specific names and one new generic name on the most tenuous of evidence, while ignoring most of the established rules of taxonomic procedure. Dr. Heuvelmans defends his proposal of the new name *Dinanthropoides nivalis* (p. 178) for the abominable snowman (the evidence being photographs of tracks in the snow and photographs of a scalp not proved to be associated with the animal making the tracks) on the incorrect ground that "this procedure is not only universal in palæontology, but happens fairly often in zoology". In support, he quotes the first description of the giant panda, the alleged Andean wolf, the takahe (*Notornis*), the pygmy hippopotamus and the okapi. In actual fact none of these were named until tangible specimens, whether skins or skulls, were examined and compared.

This bias against the professional leads Dr. Heuvelmans to turn for support for his views to writers about whose reliability and status there can be widely differing opinions. Even the introduction by the well-known author and collector of live animals, Gerald Durrell, is coloured by the same bias. Incidentally the latter's statement that the okapi was known only as an extinct relative of the giraffe until it was discovered living is quite inaccurate, and his reference to the 'surprising' discovery of the king cheetah in Southern Rhodesia is less convincing when it is known that the animal is no more than a colour mutation of the common cheetah.

In a work of this scope and length there are inevitably a number of errors and inaccuracies. A more careful checking of references would have avoided many of these. But the most serious charge that can be made is the over-emphasis on hearsay and unsupported tales which are quoted as evidence for the existence of some hypothetical animal, and the arrangement of factual evidence to support a case. For example, Fig. 81 (p. 314) is a sketch, full face, of the head of the alleged South American anthropoid or Loy's monkey, based on the photograph which is the only material evidence. In the photograph of the dead monkey the jaw sags, resulting in an unnatural appearance. In the sketch, shown with mouth shut, the face is compared with a reconstruction by von Koenigswald, 1938, of the face of *Pithecanthropus*. We are then told that if one disregards the many features in the *Pithecanthropus* face which cannot be inferred from the bones, and must therefore have been invented, the shape of the face is the same in the two. The author continues—"Although it [Loy's monkey] has been photographed full face, one can see that its profile is more like that of a capuchin monkey" (rather than a spider monkey, with which this animal is identified by primate specialists). Such ingenious presentation of evidence does not give one a great deal of faith in Dr. Heuvelmans' ability to hold a fair balance.

Again, in the case of the king cheetah (pp. 419–420), the description by R. I. Pocock in 1926 of what was thought at the time to be a very distinct new species is used as an illustration of how unreported animals can survive in a settled area for many years without discovery. But Dr. Heuvelmans does not tell the reader that in 1939 Pocock published his own conviction that this was not a new species after all, but simply a pattern mutation of the common cheetah. An illustration of the printing at full length of stories which eventually prove to have a prosaic ending is found in Chapter 20, devoted to the possibility of pterodactyls and other prehistoric reptiles still existing in Africa. Ivan T. Sanderson's vivid account of an encounter with a great black flying creature in the Cameroons (p. 491) is followed by a long discussion of its possible identity. But the footnote (p. 494) in which Sanderson is quoted (*in litt.*) as saying that the 'thing' was a bat, reduces the value of this contribution to nil.

Misquotations are frequent. The water civet, *Osbornictis piscivora* (p. 64), was discovered in the Ituri Forest not by Glover Allen in 1919 but by the collectors Lang and Chapin (of Congo peacock fame) in 1913, and it was Dr. J. A. Allen who described it in 1919. A skin and skull purporting to belong to a Nandi bear sent to the British Museum (Natural History) came not from Nyasaland (pp. 399–400) but from Uganda, hence Dr. Heuvelmans' comment on the unlikely locality has no validity. Again, we are told at the conclusion of the Nandi bear section (p. 414) that "Ivan T. Sanderson says that in the British Museum of Natural History there is a piece of fur [*sic*—'peau' in the original French edition] covered with long brown hair which may come from the animal about which armchair experts are so sceptical". If the author had checked this statement with the Museum he would have found that in fact no such piece of skin is known there.

In the class of over-confident assertions are the following. "I have shown that the Earth may still hold plenty of large unknown animals" (p. 81); "Herds of hundreds or even thousands of mammoths could easily live there [in the Siberian taiga] without running the least risk of being seen by man" (p. 350); of the alleged pygmy gorilla, named by Elliot *Pseudogorilla mayema* in a notoriously unreliable work, he says (p. 63) "The existence of this species, about which we know so little, has never been in doubt". Reference to current African primate literature would have shown Dr. Heuvelmans that, on the contrary, this name has long been relegated to the synonymy of the ordinary lowland gorilla, *Gorilla gorilla*. Do elephants, rhinoceroses and hippopotamuses really make the Earth shake, as we are told (p. 449) after reading of a Belgian in the Congo who felt a violent earthquake when a mysterious giant beast moved off unseen in the forest?

The basic natural history is often unreliable. It is not true that (p. 197), apart from species imported by man—the dingo and the rats, and several bats which could have flown across the sea—there are no wild placental mammals in Australia. There is a varied native rodent fauna, including 17 genera with 79 species, which has evolved over a very lengthy period in that continent. There are no macaques in Central Africa (p. 358). The account of the habits and distribution of the sea otter, ***Enhydra lutris***, of the North Pacific coasts, in connexion with its suggested identification as the waitoreke of New Zealand mountain waters, is largely inaccurate. The "pack of 400 seen swimming down the Californian coast in 1938" (p. 248) (presumably on their way to the Antipodes) turns out on investigation of the source quoted to refer to a much lower number of resident sea otters recovering their former status on the Californian coast, and it would be interesting to know how the "large numbers [of sea otters] along the coast of southern Chile, which seems to imply an extraordinarily long migration" reported by a quoted observer, were identified. There is a local South American coastal otter, *Lutra felina*, found commonly in those waters, which probably accounts for this confusion.

Despite these numerous criticisms, I found this book, illustrated by many photographs and sketches, very readable and sometimes stimulating. One can part company with many of the author's conclusions without denying that he has produced a useful work. He may be reassured to learn, meanwhile, that zoologists are not all quite as blind and deaf as he suggests, and that while they are waiting for some of the animals so vividly described here to appear in the flesh, they are not entirely idle. In the five years, for example, since the publication of the original French edition of this book, no less than four distinct and quite unexpected new genera of small mammals (all small enough to be beneath Dr. Heuvelmans notice) have been described from tropical Africa, and no doubt other regions will contribute their quota in due course. R. W. Hayman

MARKET STRATEGY AND ECONOMIC SURVIVAL

Strategy and Market Structure

Competition, Oligopoly, and the Theory of Games. By Dr. Martin Shubik. Pp. xviii + 387. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1959.) 64*s.* net.

IN 1944 there appeared "Theory of Games and Economic Behavior", the pioneer work by von Neumann and Morgenstern, full of promise for a uniform and dynamic theory of the firm on a new game-theoretic basis. In the subsequent fifteen years, game theory was developed and applied in many directions, but not to any effect in the theory of oligopoly and monopolistic competition, of the firm under various market conditions. What was sown in 1944 is at last bearing fruit, ample evidence being provided by this text by Dr. Shubik. It is possible to envisage now the bridging of that wide gap, disturbing to so many economists, between the traditional theory of the firm and practical business as understood by institutional economists and business schools. This is not to say that Dr. Shubik has constructed the bridge. His book is still rather severely theoretical. But he does take into account many factors which have hitherto been rather baffling—the influence of the financial strength of a firm, its technical position and its corporate form—and he has so framed his theory that empirical work can be based upon it.

Dr. Shubik achieves two quite different objectives in dividing his book into two halves. In the first part he makes a game-theoretical exposition of existing theory and throws much new light on it. In a firm under pure competition or pure monopoly, the *entrepreneur* makes his decisions on his own and economic theory has long been able to handle the situation. Under what can be described either as oligopoly or as monopolistic competition, at least one firm in planning strategy on the market for a product must take into account the influence of at least one other firm on the same market. There is then a great variety of possible market structures, according to the varying number of firms and their different situations (for example, on transport costs), according to the extent of product differentiation, and according to the relative bargaining powers or market leadership of the firms. There is competition between firms but also co-operation, coalition and collusion. This is just the kind of situation to be exploited by game theory. Duopoly as analysed by Cournot and Edgeworth is such a special case that it cannot serve for generalization. The theory of monopolistic competition as elaborated by Chamberlin and Joan Robinson is piecemeal and lacking in the essential unifying principles. Dr. Shubik succeeds in his game-theoretical approach where the traditionalist development failed.

In the second part, Dr. Shubik attempts something even more important. This is a sketch of a truly dynamic theory of oligopoly, or rather the basis on which such a theory might well be built. He allows for imperfect information among the constituent firms on a market. His starting point is the concept of a particular type of game, the game of economic survival. A game of survival or ruin is well known as an extension of an ordinary zero-sum game, applied in successive periods until one player uses all his funds and is ruined. A game of *economic* survival is again a zero-sum game as long as the contestants survive (for example, splitting up a given market) but with a bonus to the ultimate survivor. On this view, the economic activity of the firm is to maximize something, generally the stream of dividends over time, but subject to attention being paid to survival. Not all the experts in this field go all the way with Dr. Shubik in his development of the theme, but none can deny that he is pursuing with great ingenuity something very well worth while. This is an important book, not only for game-theorists, but for all economists.

R. G. D. ALLEN

THE DARK AGE IN AFRICA

Old Africa Rediscovered

By Basil Davidson. Pp. 287 + 28 plates. (London: Victor Gollancz, Ltd., 1959.) 25*s.* net.

THIS book is an attempt to accomplish the well-nigh impossible; but because the author knows a great deal about the late prehistory of many parts of Africa and can put down his knowledge in a clear and interesting manner, it is well worth perusal. It has been said that a few years ago such a work would have been impossible owing to lack of information; it can equally be said that a few years hence such a work will be quite impossible, except in the form of many volumes, because of too much data. At the same time, the reader should clearly understand the scope of the book. It is true that in the short introductories there is mention of pluvials, early lines of migrations, etc., but the author is not really concerned with very early times, his interests centring on the epoch from about 500 B.C. to the opening up of the continent by the Europeans.

As this is, perhaps, the period least understood in most African regions, the book will be very much welcomed. Of necessity the areas dealt with are somewhat scattered, but the account of the pre-Portuguese civilizations, which flourished along the east coast, and which Dr. Mathew has done so much to reconstruct, is of especial interest. Much, however, has not been included. The bulk of the rock-shelter art, at least in the Union of South Africa, falls within the author's period—indeed some of the finest of the polychromes probably date no farther back than the sixteenth century A.D. *Pace* one carbon dating, it is quite likely that the same applies to the Nachikufan culture of Northern Rhodesia at least in its later phases. This is indeed mentioned but discussed as dating to 4000 B.C. But the dating of early cultures in Africa is a notoriously controversial subject and need not be pursued further here.

It is true that the spine of the Sahara desert has been well-nigh impossible for human migration since Gamblian (last pluvial) times, but subsequent wet phases, the last one of no vast antiquity, have permitted human occupation in the fringes bordering the spine both on the north and the south, and once again much of the rock-shelter art in these regions belongs to Mr. Davidson's period. The description of the ancient town of Meroe with its great slag heaps and the Kush civilization is very well set out, and to many readers the account of Bernard Fagg's splendid pottery figurines from Nigeria which up to now have not been well published will be welcome indeed. Quite rightly, Dr. Manny's work in West Africa is noticed, and the early history of Benin is not forgotten. As has been said, the civilizations which grew up along the east coast of Africa and the (to some extent) contemporary Rhodesian buildings are treated at considerable length. The history of the early Congo is scarcely mentioned, nor is that of Angola, though the publications of the Diamond Company there have shed some light on its obscure part. The Union of South Africa, except for a brief account of Mapungubwe, is also omitted.

These gaps (geographically) are numerous, but it is difficult to see how everything could have been included in one short volume. What the author has given us is excellent reading, and he is to be congratulated on attempting what for many would have seemed impossible. It is a work which many people who care about Africa will warmly welcome.

M. C. BURKITT

IMPORTANT RAW MATERIALS

Cellulose Pulp and Allied Products

By Dr. Julius Grant. (Incorporating the third edition of "Wood Pulp".) Pp. xvi+512. (London: Leonard Hill (Books), Ltd., 1958.) 50s. net.

REFERENCE books of any size are now usually assembled by editors from chapters written by different authors—each an expert on his own subject. "Cellulose Pulp" is a survivor from an older tradition, and is the work of one author. This gives a uniformity in style, lacking in a collection of edited chapters, but anyone essaying a subject with as vast a literature as cellulose pulp sets himself a difficult task.

This book incorporates the third edition of "Wood Pulp", which was first published in 1938. Wood is quite the most important source of cellulose pulp (about 90 per cent) of the total and most of the book is concerned with the manufacture of wood pulp. The central chapters cover this well and should be of use to the general reader who is not 'highly technical' in accordance with the author's wishes expressed in the preface. Chapter 2 on history is well done, but the author could have made more of the history of wood pulp technology itself. Without wood pulp paper would have been so scarce that Forster's (Compulsory) Education Act of 1870, and similar measures in other countries, would have been useless. It is no good teaching children to read and write without adequate paper supplies, and the figures quoted by the author for paper consumption and literacy in Asia and Africa show that the problem is still with us.

Appropriately enough the main untapped source for pulp is in the tropical forests, but it is not easy to get sound information about them quickly as an enormous variety of trees grow in mixed stands and they all have different pulping properties. There is a serious sampling problem, and it is also very difficult to forecast the nature of the secondary growth, which is most important if pulping is to be put on a continuous basis.

In briefly relating his subject to cellulose technology in chapter 3 the author has run into some difficulties. The explanations of 'micelles' on page 37 and 'tracheids' on pages 49 and 50 are so short that they are misleading. Chapters 20, 21 and 22 on uses of cellulose also suffer from enforced brevity. The rayon and allied industries, and the plastics industry too, may be surprised at the small space given to their special problems. In most parts of the book sufficient references are given to enable the scientific reader to look up detailed information elsewhere, but the references to the uses of pulp are thin considering the variety of processes mentioned.

These, however, are not major issues and it is hoped that the book will be widely read for its manufacturing section. Too few people realize how important pulp technology is to our civilization.

F. LYTH HUDSON

THE CASE FOR EXTRA-SENSORY PERCEPTION

The Sixth Sense

An Inquiry Into Extra-Sensory Perception. By Rosalind Heywood. Pp. 224. (London: Chatto and Windus, Ltd., 1959.) 21s. net.

IN this volume Mrs. Heywood, an active member of the Council of the Society for Psychical Research, has collected a mixed but well-arranged selection of those cases which seem to her and doubtless to many others to offer good evidence for extra-sensory perception. She is convinced that both in England and in the United States experiments have been conducted under rigid scientific conditions which have shown that it is possible for one person to become aware of another's thoughts and feelings where such information could not in any circumstances have been acquired by sensory means.

Beginning her study with a glance into the past, she briefly summarizes the early work of the Society for Psychical Research, stressing the value of its experiments and conclusions without, however, adequately dealing with the objections to its methods which were often voiced at the time. Proceeding with her analysis she discusses the work carried out on the problem of mediumistic communications, especially those associated with the names of Mrs. Piper, Mrs. Leonard and Mrs. Coombe-Tennant. The very puzzling "Cross-correspondences" are then dealt with, and the two chapters devoted to them form an excellent introduction, although the author fails to appreciate the reason why this material has never attracted the attention that it might deserve if we knew all the facts and not only some of them. The study is concluded by a short discussion of various quantitative experiments and a short summary of some of the explanations and hypotheses that have been offered.

As a popular and necessarily brief introduction to extra-sensory perception this book is to be recommended, although always with the proviso that the facts are as stated and that further knowledge of the conditions would confirm the interpretation put upon them. It is this lingering doubt about the accuracy of the records that tends to prevent so many scientific men from taking many of the statements of psychical research workers too seriously, and this hesitation is often better justified than at first appears.

In this book Mrs. Heywood has done little to allay such doubts. What she has done is to present a further stimulating and lucid study of extra-sensory perception which may prompt independent inquirers to initiate experiments on their own account, but only after an intensive study of the subject and of the pitfalls with which it abounds.

E. J. DINGWALL

TIZARD AND THE SCIENCE OF WAR*

By Prof. P. M. S. BLACKETT, F.R.S.
Imperial College of Science and Technology, London

The Air Defence Committee

IT is sad indeed that the newly founded institute for the study of war (the Institute for Strategic Studies) should so soon have to mourn the loss of one of its most distinguished members (*Nature*, 185, 209; 1960). I wish to attempt to assess the nature of Tizard's direct personal contribution to the Allied victory in the Second World War and to trace the great influence he exerted on the general study of war, which is, of course, the explicit object of the Institute for Strategic Studies. All men's careers have much that is accidental in them, and no doubt there was an element of chance in the selection in 1934 by the Air Ministry of Sir Henry Tizard, then rector of the Imperial College of Science and Technology, as chairman of the Committee for the Scientific Survey of Air Defence. But there was none in the brilliant and decisive manner in which Tizard exploited the opportunities thus given him, at a decisive moment of world history.

The military and political outlook of the middle 1930's was indeed ominous. The world economy was just beginning to stagger uncertainly out of one of the most disastrous slumps of history; Hitler had come to power in Germany and was openly preparing for war; the advance of aeronautical science and engineering had at last gone far enough to make it possible that air power might be a vital factor in a major war—if this were so, then the extreme vulnerability of Great Britain with her high population density and her close proximity to the mainland of Europe, which only too easily might fall into enemy hands, became of major military importance. As far as there was a military doctrine of the day, it seems to have been expressed in Baldwin's famous phrase in 1933 that "the bomber will always get through".

This, then, was the background to the first meeting in January 1935 of what became universally known as the "Tizard Committee"; its official terms of reference were: "To consider how far recent advances in scientific and technical knowledge can be used to strengthen the present methods of defence against hostile aircraft". In addition to the chairman there were originally only three members: H. E. Wimperis (then director of research at the Air Ministry), A. V. Hill (physiologist at University College, London) and myself, with A. P. Rowe (later to become the director of the famous Telecommunications Research Establishment) as secretary.

During its five years of existence—the Committee was dissolved during the first years of the War—four things of major importance were achieved, mainly through Tizard's vigorous and persuasive personality. Radar, which in Great Britain originated with Robert Watson Watt at the Radio Research Laboratory of the National Physical Laboratory, was energetically supported and, with the enthusiastic support of senior airmen, introduced secretly into the Air Force. When war came in 1939 the whole east and south-east coasts of England had their radar chains operational, and this was a decisive factor in the winning of the Battle of Britain in 1940.

The second indirect but very real achievement was to bring the senior officers of the Armed Services into much closer intimacy with the research and development scientists in the Government establishments. Tizard's gifts, which will be referred to again, of inspiring confidence in young and old, in those in authority and in those at the bench or on the gun-sites: his passion for getting things done and his flair for interpreting the military requirements to the bench worker, and the technically possible to the Service chiefs: all these greatly enlivened and made more productive the scientific and technical side of the Air Force in the first instance, and then by example the other two Services.

The third achievement was the creation of mutual confidence and understanding between serving officers and university scientists; so that, when need arose, and it did arise very soon, many of the best academic research workers flocked out of the universities into radar stations, and later into Service experimental establishments, where they became a vital part of the brilliantly creative, and sometimes obstreperous, teams, whose work had so profound an effect on the waging of the War.

The fourth achievement was the recognition that scientifically trained research workers had a vital part to play, not only (as of course was traditional) in the development of the weapons and gadgets of war but also in the actual study of operations. It was due to Tizard's personal initiative as early as 1936, before, in fact, radar data on approaching aircraft were available, that civilian scientists were attached to the Fighter Station at Biggin Hill to study the art of controlled interception, as it would have to be done, when a year or two later the radar chain would become operational. This experiment seems to have been the first official recognition that the actual operations of modern war are so complicated and change so fast that the traditional training of the serving officers and personnel is inadequate. In fact, many of the operational problems which arise when new equipment comes into service require for their solution the aptitudes of the scientific research worker: for he is trained to apply scientific methods to elucidate hitherto unknown and complex phenomena.

The Biggin Hill experiment was the first step towards the fully fledged Operational Research Sections attached eventually to all the major Commands of all three Services. A further important step was taken when, at the outbreak of the War, A. P. Rowe, then in charge of radar research at Bawdsey, sent a small group of scientists to Headquarters Fighter Command to study the actual use of the radar data so as to keep the radar scientists in the establishments briefed both on the actual performance of the sets in operations and on the real operational needs of Fighter Command. Out of this group grew the Fighter Command Operational Research Section as an integral part of the Command organization. Analogous developments took place at Anti-Aircraft, Coastal and Bomber Commands, and also at the Admiralty and the War Office. These developments implied a great measure of mutual trust and understanding between the senior Service officers and the often brash and initially very ignorant scientists—ignorant, that is, of most things that went on outside a university research department. This intimate relationship between professional fighting men in charge of operations and research scientists proved highly profitable, and some of the later operations of the War were carried out under a close scientific and statistical scrutiny. The pattern of organization and method worked out in Britain during the first years of the War stimulated similar developments in the United States. Hitler's Third Reich saw no such collaboration. No doubt the almost unbroken German military successes of the first war years confirmed the highly competent military staffs in the view that they had no need to seek help from outside scientists, however brilliant. When the tide of war swept against Germany it was too late. Luckily for the Allies, Germany never produced its Tizard.

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* Tizard Memorial Lecture delivered on February 11 before the Institute for Strategic Studies.

The importance of bringing scientists into close touch with operations had been clearly foreseen during the early days of the Tizard Committee. It was well expressed by the Secretary of State for Air, Lord Swinton, then Sir Philip Cunliffe Lister. In his book "I Remember" he wrote: "There was nothing new in the use of men of science as advisors to Service Departments and to the Committee of Imperial Defence. . . . There was nothing new, therefore, in calling in science to help; but from the moment I went to the Air Ministry, I felt that in the air, with all its unknown possibilities, we wanted a much closer and more intimate relationship. The scientists who were to work with us must be from the start an integral part of the Air Staff". Of the Tizard Committee he wrote: " . . . they were at the heart of operational planning. That relationship was, and I believe always will be, the key to success in scientific co-operation".

Prominent among the senior Air Force officers who welcomed and co-operated closely with the Tizard Committee were Air Marshal Sir Cyril Newall, chief of the Air Staff 1937–40; Air Marshal Sir Hugh Dowding, Air Member for Research and Development 1930–36 and then commander-in-chief Fighter Command, and so the first operational user of radar; Air Marshal Sir Wilfred Freeman, who succeeded Dowding in 1936 as member for Research and Development; Air Marshal Tedder (now Lord Tedder), who was director-general for Research and Development during 1938–40; and Air Marshal Sir Philip Joubert, then air officer commanding Fighting Area and later commander-in-chief Coastal Command.

For the encouragement of the later developments of the application of scientific method to tactics and strategy, much credit goes to General Sir Frederick Pile at Anti-Aircraft Command, Admiral Sir George Creasy, then director of Anti-Submarine Warfare at the Admiralty, and Air Marshal Sir John Slessor, now a vice-president of the Institute for Strategic Studies. During Slessor's time as commander-in-chief Coastal Command 1943–45, the scientific analysis of the anti-submarine war became an accepted part of operational control, and statistics of air hours flown, rates of search, U-boat sightings, etc., were discussed at the fortnightly U-boat meetings at No. 10, Downing Street—once goading a senior officer to protest that wars were won by weapons and not by slide rules, so giving rise to the half-derisive but also appreciative term 'slide rule strategy'.

Unfortunately, Tizard published little about his own contribution to these developments, though no doubt much of great interest will be found in Whitehall files, when these become available to historians. So far as I know, his lecture to the Royal United Service Institution in 1946, entitled "Science and the Services", and his Haldane Memorial Lecture at Birkbeck College in 1955, entitled "A Scientist In and Out of the Civil Service", are almost all that are available: and in these, with characteristic modesty, there is little about his own achievements. So the history of these vital years has to be pieced together from personal memories.

Controversy on Priorities

Although the Air Defence Committee started up in January 1935 with only Tizard, Hill, Wimperis and myself as members, in July of the same year the Secretary of State for Air, under pressure from Mr. Winston Churchill, enlarged it by the addition of Prof. F. A. Lindemann (afterwards Lord Cherwell). It was not long before the meetings became long and controversial: the main points of dispute concerned the priorities for research and development which should be assigned to the various projects which were being fathered by the Committee. For example, Lindemann wanted higher priority for the detection of aircraft by infra-red radiation and for the dropping of parachute-carrying bombs in front of enemy night bombers, and lower priority for radar, than the other members thought proper. On one occasion Lindemann became so fierce with Tizard that the secretaries had to be sent out of the committee room so as to keep the squabble as private as possible. In August 1936, soon after this meeting, A. V. Hill and I decided that the Committee could not function satisfactorily under such conditions; so we resigned. A few weeks later Lord Swinton re-appointed the original Committee without Lindemann, but with Prof. E. V. Appleton, and a few months later with Prof. T. R. Merton, as additional members.

During the years 1935–39 the Committee investigated and assessed a large number of projects, good and bad; we visited many establishments and discussed all aspects of air defence problems with air marshals, with pilot officers and with scientists in their laboratories. Pre-eminent, of course, was radar, both ground and air-borne. The story of radar has often been told, and all I wish to emphasize is the vital part played by the confidence which Tizard did so much to build up between the senior airmen, the research workers and the responsible Cabinet ministers. The full backing of the Committee became an effective way of getting high priority given to a project. Without such mutual trust the scientific development of radar could not have forged ahead at such speed, nor would the tens of millions of pounds have been provided so rapidly and secretly by the Treasury to build the radar chain.

In 1937, Tizard was made chairman of another Committee, this time for the Scientific Survey of Air Offence, modelled on the lines of the Air Defence Committee. However, it failed to come to grips with the problems of the use of bombers and made no decisive impact. Possibly Tizard himself was too heavily involved in air defence problems easily to switch his interest: possibly the failure was partly due to the not very sympathetic response by those in the Air Force then concerned with bombing policy. Whatever the cause, I think it would be fair to hold that until the War was well advanced, Bomber Command was less scientifically minded than either Fighter, Coastal or Anti-Aircraft Command. Moreover, it was just this failure to think and analyse scientifically which delayed for too long the recognition of the extreme ineffectiveness of our early bombing offensive. Sir Winston Churchill tells how Prof. Lindemann in 1941 informed him that, of the bomber crews who thought they had found their target, it had been shown by analysing flash photographs that two-thirds were not within five miles of it. Once these facts were fully recognized, high priority was put on navigational aids of various kinds, and matters greatly improved.

Tizard's Mission to the United States

In November 1939, Tizard, then scientific adviser to the Air Ministry, suggested that A. V. Hill should go to Washington to act as scientific adviser to work with the Air Attaché, so as to get scientific help on defence problems from the United States. Since scientific developments were likely to play a major part in determining the issue of the War, the importance of attempting to get official approval for Britain to tap the vast scientific resources of the United States was very great. In a typically apt phrase Tizard described his policy as that of "bringing American scientists into the War before their Government". In this he succeeded in a startlingly successful manner. When A. V. Hill reached Washington, he realized that this objective could only be attained if a complete interchange of scientific secrets between the two countries could be arranged. With the help of the British Ambassador, Lord Lothian, President Roosevelt became convinced of the importance of such interchange, as were also the American Service chiefs. On the other hand, the task of convincing the British Government that it was useful and safe to entrust our most closely guarded secrets to the scientists and Armed Services of a then neutral country, with not a few anti-British elements in it, was not easy. But at last Tizard succeeded; and in September 1940, as the 'Blitz' on London started, the Tizard Mission, with Cockcroft as second in command, set out for the United States with the famous black box containing samples, blueprints and reports on nearly all important new British war devices. These included radar, fire control, under-water detection, aircraft turrets, Whittle's jet engine and, above all, a sample 9·1 cm. resonant cavity magnetron.

Within a few days American scientists had the magnetron working, to find it gave a thousand times the power formerly available. This remarkable invention, due to Randall and Boot working in Oliphant's laboratory at the University of Birmingham, was called by Watson Watt "a radically new and immensely powerful device which remains the heart of every modern radar equipment".

An assessment of the importance of this interchange of secret military information is given in James Phinney Baxter's book "Scientists Against Time", published in 1946 with a foreword by Vannevar Bush. The writer comments on the contents of the black box as "the most valuable cargo ever brought to our shores" and "the single most important element in reverse lease lend". He goes on: "In the early days of scientific interchange, the British gave more than they received". Later the debt was repaid many-fold; in particular, for example, by the superb centimetric anti-aircraft radar set *SCR*-584 and the proximity fuse which together led to such brilliant successes by Anti-Aircraft Command during the *V*.1 attack on England in 1944. The former set was a product of the famous Radiation Laboratory at the Massachusetts Institute of Technology, which was, in fact, founded as a result of a meeting of the Tizard Mission with American scientists in September 1940.

This imaginative act of trust, which Tizard and A. V. Hill first envisaged and finally forced through Whitehall, had immensely beneficial effects on the scientific aspects of the Allied war effort. Cockcroft reminds us that the Mission was magnificently organized by Tizard and that he had the inspiration to bring a mixed team of serving officers and scientists. For the first time our American friends heard civilian scientists discussing authoritatively the instruments of war and then heard the Service people following on with practical experience.

The Bombing Controversy

When Tizard returned to England in September 1940 on the completion of his American Mission, he worked full time at the Ministry of Aircraft Production and later became a member of the Air Council. The Air Defence Committee itself died a natural death, since an advisory committee of part-time members was not useful under fully mobilized war conditions.

There is little doubt that Tizard found less than satisfaction in what he could achieve during the years 1941 and 1942. In spite of his wide knowledge of the Government machine, he seems to have been genuinely disappointed to find that the Air Council rarely discussed strategy or tactics, but often minor administrative matters: he related that at one meeting of the Air Council the main business was the inspection of an exhibition of different designs of W.A.A.F. underwear! A deeper difficulty underlay the frustrations of this period. For it was not at all easy for a Service Ministry to incorporate a man as senior as Tizard in its taut, executive, war-time machine. On one hand he was too senior to do his own devilling among the files at the lower levels of the machine, where much of the real work of a department is done —to be sure, he was too impatient to have been very good at this—and on the other hand, he had too much self-respect to acquire influence by becoming a courtier. Tizard evidently felt that he was not wanted in Whitehall and so he accepted the presidency of Magdalen College, Oxford, where he remained, apart from some Government advisory work and several important visits abroad, until after the end of the War.

Shortly before he went to Oxford, Tizard became involved in a controversy about bombing policy which had far-reaching consequences. The interest of this story is not only its importance at the time, but also that it concerned problems of the theory and practice of bombing which, now with the advent of nuclear weapons, is giving rise to so much deep thought, subtle analysis and high controversy, carried on not least by members of the Institute for Strategic Studies.

As I remember it, what happened was this. In the early spring of 1942 the Prime Minister was extremely anxious that everything possible should be done by Bomber Command to help the hard-pressed Soviet armies at Stalingrad. About April a Cabinet paper (which was known to have been written in Lord Cherwell's office, Cherwell then being Paymaster-General and the Prime Minister's scientific adviser) was issued on the probable effect on Germany of the British bombing offensive over the next eighteen months, that is, until the early autumn of 1943. The paper laid down the policy of directing the bombing offensive primarily against the German working-class housing—middle-class housing was too spread out to be a good target and factories or military installations were too difficult to find and hit. So far as my memory goes, the paper claimed that it should be possible within the stated period to destroy 50 per cent of all houses in all towns of more than 50,000 population in Germany, if Britain concentrated all her efforts on the production of bombers and used them for this purpose. Tizard studied this paper in detail and concluded that the estimate of the number of houses likely to be destroyed in the next eighteen months was five times too high. At that time I was director of Operational Research at the Admiralty, and I was also asked to comment on the Cabinet paper. I came independently essentially to the same conclusion as Tizard—I think I estimated the error as six-fold. The main mistake made in the Cabinet paper was to assume that all bombers which would be delivered from the factories in the next eighteen months would in the same period have dropped all their bombs on Germany. The year or more from the completion of a bomber to the completion of its average operational life of twenty sorties had not been fully allowed for. The bombing survey after the War showed that the number of houses actually destroyed in the assigned period was only one-tenth of the estimate in the Cabinet paper: this agreed rather closely with Tizard's and my estimates, allowing for reduced numbers of bombers actually supplied to Bomber Command.

By this time a certain allergy to arithmetic was spreading in Whitehall, and our numerical forebodings went unheeded: the Air Ministry agreed with the Cabinet Office paper, and the policy of dehousing the German working-class population, with the object of lowering its morale and will to fight, became a major part of the British war effort.

The story goes that at that time in the Air Ministry it was said of anyone who added two and two together to make four, "He is not to be trusted; he has been talking to Tizard and Blackett". Less agreeable stories circulated: that anyone who made such calculations must be a defeatist.

So far as I know, Tizard never wrote anything of this period and spoke but sparingly; but he did comment after the War in his Royal United Service Institution lecture on the theory and practice of aerial bombing. "Experience has shown that a nation, with toughness, stamina and a will to live and work, can stand far more punishment in the form of bombardment of cities and homes than most people thought possible before the trial. No one thinks now that it would have been possible to defeat Germany by bombing alone. The actual effort in man-power and resources that was expended in bombing Germany was greater than the value in man-power of the damage caused. . . ."

If only Tizard's Offence Committee had succeeded as well as his Defence Committee, and if as a result a strong and trusted operational research section had been earlier established at Bomber Command, I think it likely that the numerical error on which the area bombing campaign was initiated would not have been made and that great advantage to the war effort would have resulted.

Whether or no this bombing controversy in 1942 was the decisive factor, there is little doubt that at that time there was not room enough at the top in Whitehall for both Tizard and Cherwell. The incompatibility of these two brilliant men, and one-time intimate friends, proved a calamity to the nation.

Too much, however, must not be made of personal factors, strong as these no doubt were. For the entry of academic scientists into fields of military strategy and tactics brings to them responsibilities for advising on courses of action on which the survival of one's country—or, to-day, in an age of nuclear weapons, the survival of civilization—may depend. So real differences of technical judgment become allied to differences of temperament and lead to lasting and often bitter struggles. The war-time controversy on bombing policy with conventional explosives and the strains that it engendered in Whitehall were but a minor foretaste of the still more bitter controversy, particularly in the United States, engendered by atomic weapons, which has left such a trail of personal antagonisms behind it.

Tizard in Whitehall, 1946–52

The result of the 1945 General Election with the consequent return of Cherwell to Oxford opened the way for Tizard to return to Whitehall. The opportunity soon presented itself, for in 1946 the new Government, headed by our President, Lord Attlee, invited him to become chairman of two new high-level advisory bodies, the Defence Research Policy Committee and the Advisory Council on Scientific Policy. Widespread acclaim from both scientists and military greeted both the formation of these bodies and the choice of Tizard to head them. Thus Tizard, for the third and last time, became a full-time Civil servant. What he achieved in the six years to his retirement in 1952 was by no means the least of his service to the nation, for there is no doubt that a great part of the credit for the growth to maturity of these two committees must go to him.

These last six years of his active life were in a real sense the fulfilment of his quarter-of-a-century-old belief in the importance to the life and prosperity of Great Britain of a close relationship between the administrative and scientific worlds, a belief which was one of the factors which led him in 1920 to forsake an academic for a Civil Service career.

It would probably be generally agreed that the Defence Research Policy Committee has proved a more effective instrument than the Advisory Council on Scientific Policy. This is certainly mainly due to the much narrower and administratively well-defined field of its activity. The executive authority residing in the Defence Ministry and the Chiefs of Staff, together with the related financial power of the Supply Ministries, gives them almost complete control of all defence matters, whether in the Government or in industry. So any advice from the Defence Research Policy Committee, on which these Government bodies are represented, is likely to be relevant and acted upon. Some of those who were intimately concerned with the working of the Committee hold that Tizard moulded it into the finest organization for co-ordinating the scientific and technical aspects of war policy which has ever existed in the Western world. During his time as chairman, from 1946 until 1952, Tizard's influence was exercised over a wide range of important and complex problems, including those arising from atomic weapons, super-sonic fighters, V-bombers and guided missiles. He shared much of the responsibility for developing the rocket range at Woomera, and when some Australians were worried that it would cost them a lot of money and perhaps no one would use it, he told them not to worry as it would attract work and men as a magnet attracts iron filings. A good example of his flair for the important practical action arose out of a discussion concerning the rocket and guided missile programme. After years of intricate debate on what the research and development programme should be, when a practical plan was eventually hammered out, Tizard ruled that the implementation of the programme should be carefully watched but that there should be no alteration or even discussion of the basic programme for two years. This ruling is reminiscent of his advice to fishermen: "Don't waste time changing flies and always know what the fishes are doing".

In his Haldane Memorial Lecture at Birkbeck College in 1955, Tizard referred to the creation of the Defence Policy Research Committee thus: "It was a revolution in organization. It was not until a short time before the Second World War started that scientists began to exercise an influence on the tactical use of weapons and later on strategy. I believe I can truthfully say that the very idea that they should interfere in such matters was repugnant to senior officers twenty years ago. It was the experience of war that caused the revolution and the appointment of the new Committee, far from being resisted by the Chiefs of Staff, was, in fact, initiated by them". "The Chairman . . . has full access to all relevant information, and attends the meetings of the Chiefs of Staff Committees not only when he is specially asked to do so but whenever he judges that his presence will be useful. No major recommendation on Defence Policy is made by the Chiefs of Staff without his knowledge and assistance".

The role of the Advisory Council on Scientific Policy, on the other hand, was much more difficult to define. Its advisory activities ranged over a wide field, including scientific and technological education and man-power, the organization of Government science and indirectly the efficiency of research and development in private industry. Taking into account the rigid autonomy of Government Departments and the privacy of private enterprise, the Advisory Council, with neither money nor executive authority, had from the beginning, and has still, a difficult task. Perhaps its main success has been in the field of scientific and technical man-power. Its relative lack of impact on the national life is not to be attributed to lack of ability of Tizard, but to the essential difficulty of injecting a small degree of direction into a social system where outside a few fields such as education and defence the word planning is not seldom used as a term of ridicule. These difficulties are currently receiving much public attention, stimulated by the vast scientific and technological power of the United States, by the much faster industrial growth of some of our European competitors and by the massive technological and economic advance of the highly directed economy of the Soviet countries. Here are problems galore for Tizard's successors on the Advisory Council on Scientific Policy to solve.

The changes that have taken place in the world of military science in the past twenty-five years, that is, since Tizard's Air Defence Committee was formed, have indeed been immense. The revolution in weapons is, of course, the most obvious, and the ultimate origin of the other organizational and psychological changes. In the 1930's it was certainly important in Great Britain to bring into the defence field from academic life as many highly trained and critical minds as possible; now in 1960 the outburst of analytical thinking and writing on military subjects by scientists, lawyers, historians, psychologists and philosophers, and, of course, by retired generals, is becoming almost overwhelming and is one of the fundamental causes of the foundation of the Institute for Strategic Studies. No serving officer could possibly find the time to read all the current literature; those amateurs like myself who write on these matters have no time to read all that the other amateurs write. The flood is greatest in the United States, where many universities have their military or foreign policy research groups or institutes, where brilliant academic minds analyse and dissect our present Western military discontents and are by no means shy of ingenious solutions. Traditionally, Britain has been averse to thinking about war in between fighting wars; once they are over we tend to forget about them until the next time. Even to-day there are only two academic posts in our universities concerned with military studies. The Institute for Strategic Studies is attempting, and I think successfully, partially to fill this gap.

This huge expansion of thinking about military subjects is, of course, largely due to nuclear weapons, and is to be welcomed as a sensible response to a highly complex and dangerous situation. On the other hand, I cannot convince myself that the practical content of the response has everywhere proved adequate to the challenge. The intellectual level of much of this discussion is of the highest, so high that I find much of it very hard to understand, and I wonder sometimes whether it is all rooted in military and political reality. The policies recommended by many of these studies seem not always to have been felt through to actual potential action. I think the essential prerequisite of sound military advice is that the giver must convince himself that if he were responsible for action, he would himself act so. During the War, when the Operational Research Group at the Admiralty had proved intellectually to themselves that big convoys were safer than small ones, before we advised the Navy to make this major change, we had to decide whether we really believed in our own analysis. I personally convinced myself that I did, by the conviction that if I were to send my children across the Atlantic during the height of the U-boat attacks I would have sent them by a big rather than by a small convoy. I think that occasionally analogous personal tests of belief in one's own recipes may be useful in the study of nuclear war. Perhaps part of the fault has been the relative silence of some professional fighting men, often may be for good Service reasons.

What I feel is needed now is more effective analytic activity within the Armed Services by those senior officers who would have, in the event of war, the actual responsibility of waging it—and conceivably of pressing nuclear triggers. All generals will agree that atomic war is much too serious a matter to be left to professors.

Tizard the Man

History has shown that Wimperis's choice in 1934 of Tizard as chairman of the famous Air Defence Committee was a very wise one: he had just the right mixture of academic and military background to enable him to build a bridge between these two very distinct worlds. Son of a captain in the Navy, who had been navigator to the *Challenger* Expedition and later Assistant Hydrographer, and who was also a Fellow of the Royal Society, the young Tizard had also been destined for the Navy but had been rejected because of eyesight. This, however, did not prevent him from becoming a pilot in the Royal Flying Corps during the First World War, where he rose to the rank of lieutenant-colonel and was awarded the Air Force Cross. Previously to this he had taken both mathematics and chemistry at Oxford, had spent a year in Berlin with Nernst, and had made researches in both physical chemistry and physics. Early in 1917 he was put in charge of all experimental flying at Martlesham Heath and later in the year went to headquarters of the Ministry of Munitions. In his lecture to the Royal United Service Institution in 1946, Tizard, commenting on his experiences at Upavon as a young Royal Flying Corps pilot, wrote of the "absence of any sign of interest on the part of the War Office . . . in the scientific development of the new fighting arm . . . as for air fighting, the enemy called the tune and we danced to it". Thus his early experiences in the Royal Flying Corps must have fixed as firmly in his mind the danger to the nation of technological inferiority in war as my experience in the Navy did in mine.

There is a well-known story of his Martlesham time. Just as he was about to take off to test a new Sopwith Camel fighter, a formation of Gotha bombers approached London. Tizard had the untested guns loaded and intercepted the bombers on their return, only to find that his guns jammed immediately. So he joined the formation and made notes about the speed and performance of the Gothas, which information was badly needed. He then waved good-bye. His most important contribution during the First World War was the development of a hitherto non-existent system of performance testing, so that afterwards no new design went into production without proper testing.

After the War he returned to Oxford and to physical chemistry, but only for a short time, as in 1920 he became assistant secretary of the Department of Scientific and Industrial Research, and secretary in 1926. Then in 1929 he became rector of the Imperial College of Science and Technology, where he remained until 1942. In 1933, he was appointed chairman of the Aeronautical Research Committee and held the post for ten years. He was thus very knowledgeable about all aeronautical matters.

With such experience and such interests it is not surprising that Tizard exploited so brilliantly the opportunity which Wimperis afforded him by making him chairman of that "grand body", as Rowe once called it. Tizard knew the Government machine well enough to realize that all committees could not achieve such great things, and in his Haldane Lecture one finds a good example of that dry wit which all his friends remember as one of his most endearing characteristics and which we all mourn to have lost. In writing of the early thirties and of the gradual growth of a machinery for scientific advice to the Government, Tizard refers to one committee in the following words: "The Committee on tsetse fly control was still sitting in 1938 without any noticeable effect on the expectation of life of these pests".

Tizard's outstanding achievement in bringing civilian scientists and serving officers into such fruitful contact was based on personal qualities of a high order. Though fundamentally of a conservative temperament and a great admirer of the virtues of tradition in national life, he was a radical in technological matters, and approached new problems and possibilities with courage, enthusiasm and originality. It is true that he often followed up too many ideas at the same time and sometimes bewildered his colleagues by alteration of course induced by the almost gad-fly quickness of mind. One reason for this was his instant readiness to look at anyone's bright ideas and to see whether they could be made to work. It is these qualities which perhaps led to him being considered a poor administrator by some people—it is true that he would sometimes chase some hare for a few weeks and then switch his interest suddenly to some more profitable quarry. However, these intellectual escapades were seldom fruitless and were an essential part of his method of spot-lighting the really important problems. When he himself had sifted the ideas thrown up by his own exuberant technical and administrative imagination he was exceedingly practical.

Throughout our long friendship, which began in 1934 when I became a member of the Aeronautical Research Committee of which he was then chairman—and what an admirable chairman he was— I have never failed to find stimulus, entertainment and enlightenment from his company. Though sometimes a little irritable, he was essentially warm-hearted and very wise. Of the individuals who have influenced me by their personality, I think Tizard comes next to Rutherford. As has already been emphasized, he had a most unusual ability to establish immediate and mutually stimulating contact with anyone doing a job; he seemed as genuinely interested in talking to a college porter, an aircraftsman or a young scientist as to a crack pilot, a professor or an air marshal. A visit with Tizard to an experimental establishment was an exhilarating experience: he left behind him a newer awareness of the tasks and a new keenness to get results.

Along with the great encouragement he so often gave to the younger men in the Services and in academic life, and his gift for inspiring them with his own enthusiasm for getting things done, his flair for setting the cat among the official pigeons, and for ignoring the normal channels, made him not always popular with the upper hierarchy. His sharp incisive wit was often exercised at the expense of the complacent, the pompous and the incompetent—among these he undoubtedly made enemies—but I have never heard of it being used at the expense of the young, the vigorous or the keen. There is a story which shows his ready wit, dating from the meeting of the British Association in Canada before the War. Tizard and a colleague inadvertently crossed over into the United States near Niagara. When challenged by a policeman, and not having their passports with them, they produced their Association membership cards. When the policeman told them that "The American Government doesn't recognize British Science", the lightning reply came from Tizard, "Oh, that's all right, neither does the British Government". As Linstead, the rector of Imperial College, recently wrote, Tizard himself played a major part in making the crack less applicable to-day.

In his Haldane Lecture, Tizard pays the highest tribute to Lord Haldane for his achievements over four decades ago, in making official circles in Britain first conscious of the national implications of the scientific-technological revolution. In a sense Haldane was the first British Minister for Science: Tizard was his disciple and we to-day are his. I will conclude by quoting to you, as expressing most aptly what many of his intimate colleagues feel about Tizard, what he felt about Bertram Hopkinson, professor of engineering at Cambridge, and one of the outstanding pioneers in Great Britain of the application of science to war, and whose death in an aeroplane accident in 1918 was a real calamity. Tizard wrote: "He had not only the position, but the qualities, to command respect from soldiers and scientists alike: and those who served under him, and who have been able to do anything for the Services in later years, have owed our own capacity largely to his leadership and inspiration".

EXPERIMENTS TO TEST EINSTEIN'S PRINCIPLE OF EQUIVALENCE

On January 30, the last day of the annual meeting of the American Physical Society in New York, three papers describing experiments designed to test Einstein's principle of equivalence were read. The principle announced by Einstein in 1911 states that a system in a uniform gravitational field is completely equivalent to a uniformly accelerated system, both for mechanical and for all other processes. This was suggested by the experimental result, which was first obtained by Galileo and more accurately by Eotvös in 1891, that gravitational and inertial masses are the same. From this principle, Einstein concluded that the rate of clocks at points of low gravitational potential is slower than at points of high gravitational potential, and deduced that spectral lines emitted from the Sun should show a shift to the red when compared with those produced on the Earth. He further deduced that the velocity of light is not constant in a gravitational field, so that light rays become curved, and that in all cases, in addition to an inertial mass, a gravitational mass $m = E/c^2$ is to be ascribed to energy.

If light travels in a system which has a uniform acceleration f, then the apparent Doppler velocity after a distance h is fh/c. Using the principle of equivalence, this implies that in the Earth's gravitational field the change of frequency of light is given by $\delta\nu/\nu = gh/c^2$. Alternatively, we may consider a description in which the rate of clocks changes in the gravitational field. Then, for the frequencies of the photon at the two points, we have $\nu_1 \approx \nu_0 (1 - \varphi_1/c^2)$ and $\nu_2 \approx \nu_0 (1 - \varphi_2/c^2)$ and $\delta\nu/\nu = \delta\varphi/c^2$, where φ is the gravitational potential, and $\delta\varphi = gh$.

It is interesting to consider the effects in a rotating system. Let us consider a system which rotates with angular velocity ω. Then a clock which is at rest in this system will be slowed down by an amount which increases as the clock is moved away from the axis of rotation. The time dilatation is given by:

$$t = \frac{\tau}{\left(1 - \frac{\omega^2 r^2}{c^2}\right)^{1/2}} \approx \tau\left(1 + \frac{1}{2}\,\frac{\omega^2 r^2}{c^2}\right)$$

where r is the distance from the axis.

An observer rotating with the system will not regard this as a special relativistic effect, since the clock is at rest in his frame; but at the clock he will observe a field of force, which he can describe by a potential, equivalent to a gravitational potential. Writing $\varphi = -\frac{1}{2}\omega^2 r^2$, we again obtain the expression $\delta\nu/\nu = \delta\varphi/c^2$.

It will be noticed that both these effects are second order in $1/c$, and are therefore extremely small. The expected shift in frequency of light from the Sun is about two parts in 10^6, and in the Earth's gravitational field, the shift amounts to 10^{-18} per cm. In a rotating system the shift can be made larger, but still not much greater than 10^{-12}, this limit corresponding to the maximum strength of materials. Repeated attempts have been made to observe the red shift in light from the Sun or from stars, but there are too many interfering processes of which little is known for convincing results to be obtained.

The situation has been completely changed by a discovery made in 1958 by R. L. Mössbauer, relating to the emission of γ-rays by nuclei in solids. It is well known that the natural width of many nuclear γ-rays, given by $\hbar/\tau$, where τ is the life-time, is extremely narrow. For example, a half-life of 10^{-7} sec. corresponds to a width of 10^{-8} volt. In a line of 10 keV., this gives a relative width of about 10^{-12}. But, in general, it is not possible to utilize this extreme resolution, because when the γ-ray is emitted, the nucleus recoils and the resulting γ-ray energy is shifted by the Doppler effect. If the nucleus were perfectly free, this would shift the line by a fixed amount; but in a substance at a finite temperature there is a large number of possible recoil velocities, resulting in a widening of the line.

Mössbauer showed that in the case of γ-ray emission from nuclei in a crystal, some γ-rays are emitted in which the recoil is taken up by the whole crystal, and therefore with a line-width which is determined by the life-time of the nuclear state. The fraction of γ-rays so emitted is given by the Debye-Waller factor, and depends particularly on the energy of the γ-ray, the elastic constants of the crystal and the temperature. It follows that when such γ-rays fall on a crystal containing the same nuclei as the emitting nuclei, there is a high probability that they will be absorbed by a resonance process. Mössbauer's original work was done with γ-rays from iridium. In this case, at 80° K., 1 per cent of the γ-rays emitted are 'recoilless'. Mossbauer showed that a relative velocity of a few cm./sec. between the source and absorber introduced sufficient Doppler shift to destroy the resonance. In September 1959, it was shown independently at Harvard and Harwell that the nucleus iron-57 emitted γ-rays showing the Mössbauer effect to a much more favourable extent. The Debye–Waller factor is in this case 63 per cent even at room temperature, and the line-width corresponds to a Doppler velocity of 0·01 cm./sec. It was realized both at Harvard and Harwell that this was a particularly suitable nucleus to use in experiments on the red shift.

The first of the three papers before the American Physical Society was read by Prof. Pound of Harvard University, and described work by himself and Mr. Rebka. He first gave an introduction to the gravitational effects similar to that given above, and showed how the effect discovered by R. L. Mossbauer offered the opportunity of testing the gravitational shift. The apparatus consists of a 400-mc. source of iron-57 mounted on a piezo-electric transducer, which is itself mounted on a hydraulic device which can provide velocities of the order of 0·0001 cm./sec. for calibrating purposes. Two counting systems were used, one a sodium iodide crystal close to the source, the other a group of crystals to obtain a large area, operated at a vertical distance of about 75 ft. A plastic bag containing helium was placed in the space between the source and the distant counters to reduce the absorption of the 14 keV. γ-ray. The transducer was operated sinusoidally, and the counts from the two counting-channels were switched into two pairs of counting systems by mercury switches, so that the pulses obtained with the source moving up were counted separately from those obtained with the source moving down. A dead time was provided to eliminate counts obtained during the switch-over period. The source on its transducer and the large counter were interchanged at intervals so that the expected sign of the asymmetry was changed. Prof. Pound showed a diagram with the asymmetries measured in the two counters in a series of two-hour runs taken in the six days previous to the meeting. The magnitudes of the asymmetries in the two counters fluctuated by larger amounts than could be accounted for by statistical fluctuations, but if averages were taken over all the runs, the measured difference in asymmetry between the counters was of the right magnitude. Prof. Pound emphasized that these results are preliminary, and that the experiments are being continued.

The second paper was read by Dr. T. E. Cranshaw and described a similar experiment performed in the Nuclear Physics Division at Harwell in collaboration with Dr. J. P. Schiffer and Dr. A. B. Whitehead. In this experiment, a 30-mc. source was mounted on a moving-coil transducer also operated sinusoidally. A gramophone pick-up was placed so that the needle was moved by the rod holding the source, and the output from this pick-up enabled the movement of the source to be monitored continuously. No dead-time was provided, the argument being that if the change-over was made when the source was moving slowly, the measured asymmetry is insensitive to the exact instant of switchover. Only one counter, a proportional counter of 6 in. diameter filled to $\frac{1}{4}$ atm. with krypton was used. Krypton is a particularly suitable gas, because it has an absorption edge just below the 14-keV. line, and so a comparatively low pressure can still give a high efficiency. Two counting channels were used, one fed from the proportional counter, and one fed with pulses from a pulse generator, and both were switched simultaneously in phase with the velocity of the source. The counts from the pulse generator were used to correct the counts from the proportional counter for inequalities of the time intervals. The results from two runs of counting were given, the source being always above the counter with an evacuated tube separating the two. In each run, cycles of four operations, including reversing the phase of the driving current in the transducer, were performed so that each part of the counting apparatus was used to count all four sets of pulses. In this way asymmetries of the apparatus were balanced out. In the first run at 12·5 metres, 30 cycles of 8 hr. each were carried out, and the asymmetry obtained corresponded to a Doppler velocity of $3\cdot9 \times 10^{-5}$ cm./sec. In terms of the expected shift, this gives the result 0·96 ± 0·43. The error quoted is that given by statistical fluctuations, and a χ^2 test on the separate 8-hr. runs showed deviations not appreciably greater than could be expected from the statistics. The second run at 3 m., consisting of 8-hr. cycles, was performed as a somewhat redundant test to ensure that the cycling procedure did indeed eliminate systematic errors. The average of these runs was of the expected magnitude, but the fluctuations in the individual points were about twice as large as those expected from statistics. This was attributed to the effects of the increased counting rate with the source at such a short distance from the detector. These experiments will also be continued with a stronger source to obtain a more precise result.

The third paper was read by Dr. J. P. Schiffer, who is a visitor at Harwell on a Guggenheim Fellowship from the Argonne National Laboratory, and described an experiment also carried out at Harwell on a rotating system in collaboration with Drs. H. J. Hay, T. E. Cranshaw and P. A. Egelstaff. The apparatus consisted of a source of iron-57 deposited on an iron cylinder 0·8 cm. in diameter, clamped rigidly between two circular 'Dural' plates of 6 in. diameter. At a radius of 6·6 cm. near the circumference of the plates, a 'Perspex' strip was fixed carrying an iron-57 absorber. The whole system was rotated at speeds up to 500 cycles per sec., giving accelerations of 65,000 times terrestrial gravity. Any shift in the energy of the γ-rays would be observed as a decrease in their absorption, that is, by an increase in the counting rate. The observed change in counting rate as a function of angular velocity followed the variation predicted by the theory of relativity with approximately 10 per cent accuracy due to counting statistics.

The two types of experiment together constitute an illustration of the principle of equivalence. The first type is the only one involving real gravitational fields and this by itself constitutes a test of a prediction of this principle. The rotor experiment can be understood within the framework of special relativity alone. It is, however, the first experimental test of a relativistic effect in a rotating, and therefore accelerated, system of co-ordinates. Theoretical physicists are not completely agreed about the significance of this latter experiment.

T. E. CRANSHAW
J. P. SCHIFFER*

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* John Simon Guggenheim Fellow, on leave from Argonne National Laboratory, Lemont, Illinois.

OBITUARIES

Dr. Oliver E. Buckley

Dr. Oliver E. Buckley died on December 14. He will be remembered for many things in the science and technology of electricity. His early years were notable for researches on such diverse topics as the Hall effect, ionization manometers, magnetic materials and submarine cables. His later years were occupied with important government advisory committees and boards. In between came an outstanding but quietly accomplished achievement that will increase in stature as the years go by. This was his work in assuring that basic research will have a secure place in the industrial laboratories of the future. It was he who took over from Dr. F. B. Jewett, the first president of the Bell Telephone Laboratories, a group of scientists who had already given promise of what basic research could do for an industry. Dr. Buckley saw and appreciated this promise. It was he who protected this group from pressures to digress into development. It was he who made it into a recognized function of the Laboratories' activities. It was he who turned it over in mature form to his successor, Dr. Mervin J. Kelly, when he retired from the presidency of the Laboratories in 1951 and as chairman of the Board in 1952.

Dr. Buckley was born in Sloan, Iowa, in 1887. He received the B.S. degree from Grinnell College in 1909 and the Ph.D. from Cornell University in 1914. He immediately joined the Western Electric Company as a research physicist. He soon developed an ionization manometer which was an important tool in the early work on vacuum tubes. Taking an interest in new magnetic materials, he was quick to recognize possibilities in the use of permalloy to increase the speed of telegraph transmission on submarine cable. When perminvar came along, he envisaged a submarine cable capable of transmitting human speech. This project was carried far enough to show that it was quite practical from a technical point of view. The economics of the times determined its fate. Dr. Buckley's interest never flagged, however. In 1932 he started a group working on a multichannel system employing repeaters powered at relatively low voltage with current sent through the cable itself. While this project was never completed in the original form, the work done on it was basic to the cables that now are in operation carrying many conversations across the Atlantic, to Alaska, to Hawaii and soon across the entire Pacific.

When the Western Electric Company's engineering department became Bell Telephone Laboratories in 1925, he was appointed assistant director of research. In 1933 he became director of research and in 1936 executive vice-president. He served as president from 1940 until 1951 and as chairman of the Board until he retired in 1952. During this time many technical advances were made. These included the wave-guide, the transistor, information theory and applications including plastic cable sheaths, automatic message accounting, nation-wide dialing, and micro-wave relay networks.

In the course of his career, Dr. Buckley received many honours and served on a number of important committees. He was a member of the General Advisory Committee of the Atomic Energy Commission, 1948–54, was appointed by the President to be the first chairman of the Science Advisory Committee, and was a member of the National Inventors' Council. He received the Medal for Merit, the nation's highest civilian award, in recognition of his contribution during the Second World War. He was a Fellow of the American Physical Society, the American Association for the Advancement of Science, the Acoustical Society of America, the American Academy of Arts and Sciences, and the American Institute of Electrical Engineers, of which he was vice-president during 1946–48. He was a member of the U.S. National Academy of Sciences, the New York Academy of Sciences, the Franklin Institute, and the American Philosophical Society, of which he served as vice-president in 1954–55.

In the face of these and other achievements, honours and activities, Dr. Buckley retained his characteristic friendly enthusiasm and love for people. He knew a great many of them, and knew how to work with them. To-day, more and more, we see industrial laboratories engaged in basic research. A large share in promoting this belongs to Dr. Buckley. By example in his own laboratories, by logic and inspiring discussion on boards and in committee meetings, and by personal contacts he made people see the need for this type of research and the harvests to be reaped from its cultivation. He has sowed well. The reaping will increase with the years.

F. B. Llewellyn

Captain Guy Gardner

Had he lived a little longer, Captain Guy Gardner's name would have been better known to archæologists in Great Britain than can be the case before the important record, in book form, of his prolonged excavations at the great proto-historic site of Mapungubwe in the Transvaal is available. His death in Johannesburg on Christmas Eve, aged seventy-eight. emphasizes the evils of over-delay in publication, in this case beyond the author's control. For the work was done in six consecutive seasons between 1935 and 1940, and written up during and after the ensuing war years. Its speedy publication is surely now a duty imposed on academic bodies in the Union of South Africa with or without a Government subvention.

Gardner was not a professional archæologist, and, indeed, had his first taste of field-work with me in the Faiyum in 1927–28. But though farming in Natal had been his livelihood ever since he settled in South Africa as a young man, he had loved history and archæology, particularly classical, all his life ; and in the Graeco–Roman remains of my Faiyum concession he rejoiced in his introduction to the reality of the beloved. "The Desert Fayum", Chapter 31, contains his unaided report, illustrated by plans and by drawings of pottery which at the time were the first substantial groups known securely dated to the reign of Ptolemy II.

A more ambitious opportunity followed. The now-famous kopje site of Mapungubwe Hill, accidentally discovered in 1932, had been partially dug by the late Neville Jones and J. F. Schofield in 1934 ; it had disclosed an iron-age Bantu culture, rich in gold ornament, yet the work of predominantly non-negroid people. This presented unresolved archæological difficulties of interpretation and date manifest in the ensuing publication, "Mapungubwe: Ancient Bantu Civilization on the Limpopo".

Continued research was obviously imperative. General Smuts in a London meeting urged my acceptance of the task; but my reply was to the effect that, on Zimbabwe experience, I preferred passionless archæology. South Africa, I felt, should be encouraged to produce its own excavators. The choice fell on Guy Gardner, who devoted unreservedly his time and energies to six seasons of arduous work, with only occasional European help. Were it not for his letters home, and for his published presidential address to the South African Archæological Society in 1958, and articles in the *South African Archæological Bulletin*, little would be known of his results or his opinions upon them; and even so, obscurities abound for lack of detail. In bare outline we know of his examination of cemeteries and deeply piled middens at the foot of a kopje named Bambandyanalo adjacent to Mapungubwe Hill. The culture, characterized by fine pottery and unusual burial ceremonials, is not that of Jones's and Schofield's people on Mapungubwe, though both populations fall into the Boskopoid–Bush category of South African anatomists, which Gardner terms 'Proto-Hottentot'. His excavations, when published, will have the great advantage over the earlier ones in that carbon-14 samples will introduce fixed dates (within their own wide margins of uncertainty) both for the 'gold users' of the Hill and for the less opulent but more interesting people of Bambandyanalo, who may be at least a thousand years old.

Guy Gardner's meticulous methods of excavation and record, graphic as well as verbal, are a guarantee that a mine of hitherto unknown information awaits impatient readers of his *magnum opus* to be.

G. CATON THOMPSON

NEWS and VIEWS

Physiology at Bedford College, London :
Prof. Margaret Murray

THE retirement of Prof. Margaret Murray from the chair of physiology at Bedford College prematurely because of ill-health is a great misfortune to many. She has been associated with Bedford College for forty-two years, as a student when she gained first-class honours in physiology and as demonstrator and as lecturer in biochemistry until in 1947 when she became professor and head of the Department. More than anyone else she has been responsible for the steady progress of physiology for science students in London, and the University owes her much for her services. She has been not only a member of the Boards of Studies in Physiology, and of Biochemistry, but also served on the Board of the Faculty of Science and on innumerable special committees, to which she always gave most conscientious attention. She was chairman of the Special Advisory Board on Dietetics, Nutrition and Household Service for many years and was a member of the Committees of the Medical Research Council on Dental Diseases and on Goitre. She has been a member of the Committees of the Physiological and Biochemical Societies and on the Editorial Board of the *Journal of Physiology*. Her opinions, if sometimes dogmatic, were clear, unafraid, to the point and always respected. Prof. Murray has a wide field of interest on the biochemical side of physiology. Her researches have earned her a high reputation in the scientific world, for her painstaking accuracy is known to all. Her intercollegiate classes on "Blood" were a model of what such classes should be, involving as they did an enormous amount of preparation. In later years she has taken a special interest in the prevention of dental decay by fluorine and its possible hazards, and in the prevention of goitre in hot countries by the use of sodium iodate. In 1937 she was awarded the D.Sc. of the University and now she has been granted the title of professor emeritus.

Prof. W. F. Widdas

DR. W. F. WIDDAS, reader in physiology in King's College, London, has been appointed to succeed Prof. Murray. He qualified in medicine in Durham in 1938 and after a short time in practice volunteered for the Royal Army Medical Corps. He served with the British Expeditionary Force until Dunkirk and later held staff appointments at Northern Command and at the War Office with the rank of major. In 1947 he joined the staff of Prof. A. St. G. J. Mc. Huggett at St. Mary's Hospital Medical School, where he worked with a team on fœtal physiology and rapidly became senior lecturer. Dr. Widdas's special interest has been the physico-chemical aspect of physiology, with special reference to the transfer of substances across animal membranes, and he has made notable contributions in this field. He was awarded the Ph.D. (London) in 1953 for a study of the kinetics of glucose transfer into the red cells of adult and fœtal animals, and in 1958 the D.Sc. for his further researches. In 1955 he moved to King's College, where he became reader in 1956. There he has not only been a very active teacher of large classes of medical and science students but has also conducted a very popular intercollegiate course in cell permeability for the B.Sc. (special). He has continued his studies on the red cell and by a most novel method extended his work to cardial muscle, which he has shown loses water during contraction. It is of interest that Dr. Widdas is the twelfth member of the staff of the Department of Physiology of King's College to become a professor in recent years.

Scientific Department of the National Gallery :
Mr. F. I. G. Rawlins, C.B.E.

MR. F. I. G. RAWLINS, scientific adviser and deputy keeper at the National Gallery, will retire at the end of March after twenty-five years service. After graduating at Cambridge (Trinity College) and research at the University of Marburg, he held a number of posts at Cambridge where he was also engaged in research in infra-red spectroscopy. He was the first vice-president and secretary-general of the International Institute for the Conservation of Museum Objects, and is technical director of the Central Council for the Care of Churches and a member of the Cathedrals Advisory Committee. He was the first scientific adviser to the National Gallery, and from very small beginnings he has built up a department which has made notable contributions in its special field. Mr. Rawlins was responsible for the arrangements for the transport of the pictures from the Gallery at the outbreak of war and for providing them with a place of refuge in a quarry in Wales where they were stored under carefully controlled conditions of temperature and humidity. He was successful in securing a grant from the Nuffield Foundation for scholarships at the Gallery. The scholarships proved of such value that the Treasury has assumed responsibility for these posts. The research that has thus been initiated by him at the National Gallery and carried on under his direction may well lead to fundamental improvements in methods of conservation. Mr. Rawlins was made C.B.E. in this New Year Honours.

Mr. R. H. G. Thomson

MR. R. H. G. THOMSON, who succeeds Mr. Rawlins at the National Gallery, has been research chemist (senior scientific officer) there since 1955. Born in 1925, he was educated at Charterhouse and Magdalene College, Cambridge, where he obtained a first-class in both parts of the Natural Sciences Tripos. He stayed on to read English, before joining Dr. Charles Singer in the preparation of "A History of Technology" to which Mr. Thomson was himself a contributor. He is a *rapporteur* of the Scientific Laboratories Committee of the International Council of Museums, and presented a paper at its Copenhagen meeting in 1959 entitled "Picture Varnishes To-day". This aroused considerable interest, and marked substantial progress in a highly specialized field. It is a major theme for further fundamental research in the National Gallery Laboratory. Mr. Thomson has recently become editor of *Studies in Conservation* (the journal of the International Institute for Conservation of Historic and Artistic Works) which brings him into touch with a wide circle of investigators, both in Great Britain and abroad. He is assured of the best wishes of friends and colleagues as he enters upon his new responsibilities.

Geological Society of London Awards

THE following awards for 1960 of the Geological Society of London have been announced: *Wollaston Medal* to Prof. C. E. Tilley, professor of mineralogy and petrology in the University of Cambridge, for his distinguished researches in mineralogy and petrology especially with relation to metamorphism; *Murchison Medal* to Dr. A. G. MacGregor, formerly assistant director of H.M. Geological Survey (Scotland), for his work in vulcanology and petrology and problems of the metamorphic rocks in Scotland; *Lyell Medal* to Dr. Doris Reynolds for her work on the rocks of Northern Ireland and her contributions to petrogenesis; *Prestwich Medal* to Sir Vivian Fuchs in recognition of the geological work of the expeditions which he has led in many parts of the world; *Wollaston Fund* to Mr. M. A. Calver, of H.M. Geological Survey, for his work on faunas of the Coal Measures, especially of the North of England and the Midland Valley of Scotland; *Murchison Fund* to Dr. D. V. Ager, lecturer in palæontology in the Imperial College of Science and Technology, for his researches on brachiopods and on Mesozoic stratigraphy; a *moiety of the Lyell Fund* to Dr. J. D. Lawson, lecturer in geology in the University of Birmingham, for his work on the stratigraphy and palæontology of the Ludlow rocks of the Welsh Borderland; *another moiety of the Lyell Fund* to Dr. R. H. Cummings, lecturer in geology in the University of Glasgow, for his researches on the morphology and evolution of the Carboniferous Foraminifera.

British Scientific Attachés

IN a written reply to a question in the House of Commons on February 3 regarding scientific attachés, the Minister of Education, Sir David Eccles, as representing the Minister for Science, said that five scientific attachés are now attached to British embassies abroad. The attaché at Washington is accredited also as scientific adviser (civil) to the High Commissioner in Ottawa. The attaché at Paris is accredited also to Berne and carries out liaison duties in Italy and the Benelux countries. The attaché at Bonn is accredited also to Vienna. The attaché at Stockholm is accredited also to Copenhagen and Oslo, and carries out liaison duties in Finland. In addition, a scientific adviser to the High Commissioner for the United Kingdom is stationed in Delhi. Attachés or advisers are thus accredited to eleven United Kingdom embassies or High Commissioners' offices.

Appointment of Scientific Officers for United States Embassies

THE U.S. Department of State has announced the appointment of seven additional scientists for its science programme. Three of those appointed are assigned as science officers in New Delhi, Buenos Aires and Rio de Janeiro, four as deputy science officers in London, Stockholm, New Delhi and Tokyo. The men selected are: Earnest Watson, dean of the faculty, California Institute of Technology, as science officer for New Delhi; Neal Weber, professor of zoology, Swarthmore College, as science officer for Buenos Aires; Harry W. Wells, chairman, Upper Atmospheric Section, Carnegie Institution of Washington, as science officer for Rio de Janeiro; John B. Bateman, United States Army Chemical Corps, as deputy science officer for London; William Littlewood, United States Navy Hydrographic Office, as deputy science officer for Stockholm; David C. Rife, International Co-operation Administration Adviser to the Government of Thailand, and formerly professor of zoology, Ohio State University, as deputy science officer for New Delhi; and Marshall Crouch, professor of physics, Case Institute of Technology, as deputy science officer for Tokyo. This is the second series of appointments to be made for the science programme of the Department of State under Wallace R. Brode, science adviser to the Secretary of State. The first appointments assigned science officers to U.S. Embassies in London, Paris, Rome, Bonn, Stockholm and Tokyo (see *Nature*, 183, 15; 1959).

Anglo-American Space Research

QUESTIONED in the House of Commons on February 15 regarding the Government's future policy towards space research, the Minister of Aviation, Mr. Duncan Sandys, as representing the Minister for Science, said that the plans for Anglo-American co-operation referred to in the Prime Minister's statement of May 12 and of the Minister of Supply's statement of July 29 were developing satisfactorily and we were proceeding with the preparation of the instruments to be carried in the first satellite. Vertical soundings and experiments in the ionosphere were being carried out in co-operation with Canada and Australia and all Commonwealth countries had been invited to give suggestions on all aspects of the programme and were being kept fully informed of progress. Britain was also going ahead with design studies of the modifications which would be needed to allow Blue Streak and Black Knight to be used for launching satellites and meanwhile Britain would make any use she could of test firings to obtain further information about conditions in the upper atmosphere.

U.S. Planning Committee on Planetary Sciences

The Executive Committee of the American Geophysical Union of the National Academy of Sciences has formed a Planning Committee on Planetary Sciences. The purpose of the new committee will be to accommodate the increasing number of American Geophysical Union members engaged in planetary and space research by expanding the Union's activities in this field. The following committee officials are announced: Homer E. Newell, jun. (deputy director, Office of Space Flight Programmes, National Aeronautics and Space Administration) as chairman; Robert Jastrow (Theoretical Division, Goddard Space Flight Center) as secretary. Other members of the committee are Philip H. Abelson, Leroy R. Alldredge, J. W. Chamberlain, Thomas Gold, J. G. F. MacDonald, Hugh Odishaw, Allen Shapley, E. H. Vestine, Harry Wexler and Charles A. Whitten.

School Children in Africa

More than half the world's children are now in school. Unesco figures, collected over a three-year period, estimate that there are approximately 550,000,000 children aged 5–14 years in the world to-day, but only 300,000,000 of them are in school. During the early 1950's, the world passed the half-way mark in getting school-age children into classrooms. The enrolment figure was 48 per cent in 1950 and 55 per cent in 1954. Many countries to-day still show low enrolment figures, especially in Asia and Africa; but it is in these countries that enrolments are increasing the most. In the Belgian Congo, for example, the number of primary school pupils rose from 145,000 in 1930 to 1,060,000 a quarter of a century later (*South Afr. J. Sci.*, 55, No. 7; July 1959). Enrolment in Ghana rose from 55,000 in 1932 to 508,000 in 1954. Girls' education is no problem in Basutoland where the difficulty lies in getting more boys into school. Owing to the economic pattern of Basuto life, where boys are traditionally herdsmen, girls outnumber boys in class by two to one.

Contemporary Physics

Two numbers of the new journal, *Contemporary Physics*, have so far appeared, and there are excellent articles in both (Taylor and Francis, Ltd., 18 Red Lion Court, London, E.C.4. 27*s*. (4 dollars) per vol. of six parts; 5*s*. (75 cents) per part). The journal aims at interpretation and review—to maintain the highest level of scientific content, but at the same time to present the content in such a way that the young graduate, and the teacher needing a refresher course to bring him up to date with modern developments, may understand without undue difficulty. The editor has succeeded in obtaining first-rate authors to write articles on interesting and topical subjects, but generally the standard is rather high and the text too burdened with fearsome mathematics and frightening graphs. There are exceptions, of course, like the articles on skidding friction by Barbara Sabey, of the Road Research Laboratory, and on diamond by Prof. S. Tolansky; but the reader turning with expectation to Sir Harrie Massey's article on space research may well be put off by the fact that it contains no illustrations at all. It is conceivable that it will be the university teacher and the more advanced physicist who will spend many enjoyable hours reading *Contemporary Physics*. The article, "Molecular Beams", by Prof. O. R. Frisch, for example, is an excellent introductory survey to the subject which will neither dismay a postgraduate research physicist working in some other field of physics, nor give him the impression that the author is talking down to his reader.

Specific articles in both numbers are devoted to historical or educational aspects of physics. In No. 1 (October 1959), Dr. C. Kellner discusses Alexander von Humboldt and the organization of international collaboration in geophysical research; Mr. H. V. Beck, the selection and design of experiments for alternating current teaching; and in No. 2 (December 1959), F. Lester describes a course in the physical sciences for teachers in training; Sir John Cockcroft, the newly founded Churchill College; and L. R. B. Elton the vexed question of a clearing-house to help students to find admittance to honours courses in physics. *Contemporary Physics* deserves to do well; perhaps, without departing too much from its initial standard, its content could be made more appealing in form and less suggestive of another learned society journal, so that it may reach the wider public for which it is intended and which it richly deserves.

Botanica Marina

Scientific periodicals are already so numerous that the appearance of a new one inevitably arouses misgivings. There does, however, seem to be a very good reason for the birth of *Botanica Marina*, the first number of which has recently appeared (Cram, de Gruyter and Co., Hamburg, 1959). As Prof. Drescher-Kaden points out in the introduction, classical botany has for hundreds of years been essentially *Botanica Terrestris*, but recently both planktonic and benthic marine algae have been attracting more attention than formerly. Their intrinsic interest is great and their practical importance large and increasing, so it is obviously desirable to have a journal for the publication of work on them. The editorial committee, which is truly international, must inspire faith in the future of this journal, while the contents of the first part give an indication of the range of studies to be included—cell physiology, chemistry, statistical ecology and what might be called 'straight phycology'.

It would be interesting if in some future number we could be given an account of the debt botanists owe to the work of zoologists and chemists in the investigation of marine phytoplankton. It must be considerable. *Botanica Marina* looks like becoming an essential periodical for every library with marine interests and a useful one for such others as can afford it.

Technological Education and the Nuffield Foundation

The Nuffield Foundation has made an imaginative and constructive approach to senior colleges of technology which should do much to raise the standards of technological education in Great Britain. In a letter to the principals of those colleges which provide advanced sandwich courses, the director, Mr. Leslie Farrer-Brown, has indicated that the Foundation would be prepared to consider any requests for financial help which would lead to improvement in the quality of technological education provided by the colleges. The Foundation in no way seeks to limit the way in which senior colleges of technology will develop and hopes that each will grow in a manner in which it can best serve the needs of industry. As education for technology must be given parity in technical institutions and in industry, there is room for considerable experiment about the ways in which this should be organized. So, too, with the teaching methods which have yet to be evolved. Among a number of tentative proposals, Farrer-Brown indicates that "perhaps, in suitable cases, the attainment and maintenance of high standards of teaching and the complementary training in industry, might be fostered if teachers of their colleges and their associated educational staffs in industry had the opportunity to study educational methods at technological institutions overseas". In the first place principals have been invited to submit informal proposals.

Leverhulme Trust

THE second report of the Leverhulme Trustees classifies the awards made during the first twenty-five years of the activities of the Leverhulme Trust Fund (The Leverhulme Trust. Analysis of Grants 1932–1955: Second Report. Pp. 18. London: The Leverhulme Trust, 1959). Of the total of £854,358, £179,400 was awarded to universities and centres of higher education; £84,000 to learned societies, research institutions, etc.; £132,700 to schools; £3,600 to archæology; £54,000 to the arts; £28,300 to architecture and town planning; £115,300 to medical research; £58,600 to trade charities; £26,400 to trade educational grants; £40,800 to miscellaneous grants, with £131,258 on the Advisory Committee, individual research awards and grants. No capital grants or contributions are made to appeals of a general nature. Long-term grants are, as a rule, excluded, and broadly speaking the Fund goes mainly to the furtherance of research in the universities, teaching hospitals and other centres of higher education, the Trustees' primary concern being to foster new ideas which may lack support from other sources.

Scientific Research in Schools

THE Committee on Scientific Research in Schools which was established by the Council of the Royal Society on March 7, 1957, reports that interest shown by schools in undertaking research projects has increased and all the research projects originally supported by Shell Research, Ltd., have now been transferred to the Committee. There are now forty-eight schools undertaking scientific research; some of the schools have more than one project under way and a total of fifty-seven separate research projects are now being administered compared with twenty-five projects last year. Various requests for advice have also been received from another two schools and these are under consideration. During the year a sum of more than £2,000 has been granted to those carrying out research. The Council of the Royal Society has provided a sum of £2,000 and Shell Research, Ltd., have given an additional £127 10*s*. in addition to their support of research in two schools. A scheme has also been arranged with the Director of the National Physical Laboratory, whereby, during the summer holidays, schoolmasters selected by the Science Masters' Association can visit the Laboratory and carry out some scientific work there. All research projects with which the Committee is dealing are carried out with the specialist advice and assistance of Fellows of the Royal Society and by others who have agreed to act as advisers. The Committee has been stimulated by the continued and growing response to its scheme and looks forward both to receiving further requests from schoolmasters and schoolmistresses and to the continued support of the Fellows of the Royal Society and others in acting as advisers and assistants in the extension of its activities. This enterprising scheme should do much to raise the standard of science teaching in schools.

The Geological Survey of Great Britain

THE Summary of Progress of the Geological Survey and Museum for 1958 records much work in the field and offices. With regret, however, it directs attention to the shortage of trained cartographical draughtsmen, who are so important in the preparation of the publication of geological maps. This regret is indeed shared by many who have to wait for such long periods to see the results of the field and subsequent reports that have been prepared by the geologists. Primary survey has been carried out in several areas and revision was continued mainly in areas of economic importance, particularly the coalfields. Statistics of mapping are given for the quinquennium. Exploratory boring is now an important part of the work of the Survey and the results of various geophysical methods are interesting. The Survey has continued as geological consultant to the United Kingdom Atomic Energy Authority, and an airborne radiometric survey of the remainder of Cornwall, much of Devon and part of Somerset has been undertaken. The Museum attracted nearly 390,000 visitors, and re-organization of the reserve mineral and economic collections and research upon them continued.

A Technique for Capturing Birds and Mammals

CAPTURING large numbers of wild birds and mammals for the purpose of marking the animals for ecological and behavioural studies is usually a difficult task. A paper by Ronald F. Labisky describes a technique which was found effective for capturing pheasants (*Phasianus colchicus*) and certain other animals by using bright lights at night to blind them temporarily ("Biological Notes", No. 40. Natural History Survey Division, University of Illinois). The capturing of pheasants by night-lighting, also called 'jack-lighting' or 'shining', has been employed in practical game management since the late 1920's. Oscar Johnson reported that workers in South Dakota captured about 10,000 pheasants during the winter of 1926–27, and 12,000 pheasants during the winter of 1929–30, by 'shining' roosting birds with motor-car headlights. More recently, workers in Idaho, South Dakota, Nebraska and California have employed night-lighting in capturing pheasants. In Illinois, one of the problems in pheasant research has been that of capturing and marking a large enough number of wild pheasants to permit investigations of behaviour, population dynamics and movements. While bait trapping of wild pheasants is successful in many States, it is difficult, or frequently impossible, in east-central Illinois during autumn and winter; this difficulty is probably the result of an abundant year-round food supply and moderate winter weather.

In order to facilitate the pheasant research programme in east-central Illinois, a mobile night-lighting rig was designed. The night-lighting apparatus was also found to be useful for observing and studying the behaviour of animals during the hours of night. This study technique is particularly valuable when many animals have been marked for individual recognition.

Stone-fruit Viruses

In serological and physical studies of stone-fruit viruses, R. S. Willison, M. Weintraub and J. H. Tremaine have reported that strain *Y*.4 of the cherry yellows virus was purified from cucumber by four methods (*Canadian J. Bot.*, 37, No. 6, 1157 ; 1959). These differed mainly at the clarification stage. Two of the methods, which are given in detail, yielded infective preparations. The virus was usually inactivated by freezing. Ultra-violet absorption spectrum analysis, and particle-size and sedimentation-rate determinations, supported by infectivity tests, indicated that, in these infective preparations, the virus replaced most if not all of the components found in comparable extracts from healthy sources. The agar-gel diffusion technique was used for precipitin tests. Virus antigen from cucumber sources reacted with homologous antiserum or with antiserum derived from infected cherry petals and vice versa.

Letters written by Frédéric or Irène Joliot-Curie

P. Biquard, writing on behalf of the Association Frédéric et Irène Joliot-Curie, Institut du Radium, 11 rue Pierre-Curie, Paris 5e, asks if readers of *Nature* possessing letters written by Frédéric or Irène Joliot-Curie would inform him. It is one of the aims of this Association to publish such correspondence, and it would like to receive such letters, or photocopies. The letters will only be published if their owners give the necessary authorization.

Classical Languages in Oxford Entrance Requirements

Congregation at Oxford has approved a scheme proposed by the Committee for Admission to the University for new entrance requirements. The position of Latin and Greek was the topic of widest interest in the new proposals, under which Latin would no longer be essential. The Committee for Admission recommended exemption from Latin for those who have a pass at Advanced Level in a General Certificate of Education examination or a college award in mathematics or science. This proposal was accepted, and a statute embodying the decision will be brought before Congregation next term. To become effective, the statute will require a two-thirds majority.

Colombo Plan Appointments

In a written answer in the House of Commons on February 12, the Minister of State for Commonwealth Relations, Mr. R. Thompson, stated that in consultation with the Government of India, the United Kingdom Government is arranging to recruit and provide, under the Technical Co-operation Scheme of the Colombo Plan, ten members of the professorial staff of the College of Engineering and Technology in the University of New Delhi, and two science workshop supervisors. It is intended that the professorial appointments should be for average periods of five years.

Announcements

Sir Harry Melville, secretary of the Department of Scientific and Industrial Research, will formally open the new laboratories of the Coil Spring Federation Research Organization in Sheffield on March 24 (see *Nature*, 184, 777 ; 1959).

The twenty-fourth meeting of the European Federation of Chemical Engineering will take the form of an International Symposium on Distillation, which will be held in Brighton during May 4–6. Further information can be obtained from the Institution of Chemical Engineers, 16 Belgrave Square, London, S.W.1.

Under the auspices of the Laboratory Animals Centre, a symposium on the Provision of Animals for Cancer Research will be held at the Royal Society of Medicine on May 23. Further information can be obtained from the Secretary, Laboratory Animals Centre, Medical Research Council Laboratories, Woodmansterne Road, Carshalton, Surrey.

The Department of Mechanical, Civil and Chemical Engineering, Royal College of Science and Technology, Glasgow, is organizing a symposium on Nuclear Reactor Containment Buildings and Pressure Vessels during May 17–20. Further information can be obtained from the Secretary of the Royal College of Science and Technology, Glasgow, C.1.

The Royal Microscopical Society is arranging a two-day meeting in conjunction with the Electron Microscopy Group of the Institute of Physics, to be held in Leeds on March 31 and April 1, to discuss "Co-ordination of Light and Electron Microscopy". Application forms can be obtained from the Secretary, Royal Microscopical Society, Tavistock House South, Tavistock Square, London, W.C.1.

The seventh international congress of the European Ceramic Association is to be held in London during May 23–28. The purpose of the conference is to establish opportunities for the discussion of common interests and problems between specialists from all parts of western Europe. Further information can be obtained from the organizing secretary, Mr. H. O. Cox, 36 Victoria Street, London, S.W.1.

The Chemical Society, the Institute of Petroleum, the Royal Institute of Chemistry and the Society of Chemical Industry are jointly organizing a symposium to be held in the Chemistry Lecture Theatre, the University of Manchester, on March 24, on the Industrial Chemistry of the Lower Olefines. Further information can be obtained from Dr. P. J. King, Department of Chemical Engineering, Manchester College of Science and Technology, Sackville Street, Manchester 1.

Dr. Marie Boas's book "Robert Boyle and Seventeenth Century Chemistry" (Cambridge University Press, 30*s*. net ; see *Nature*, 183, 850 ; 1959) has been awarded the Pfizer Prize of the History of Science Society for the best book on the history of science by an American author published in 1958. "The Rotation of the Earth", by Prof. W. Munk and Dr. G. J. F. MacDonald of the University of California, has been awarded the Monograph Prize of 1959 of the American Academy of Arts and Sciences for a work on a physical or biological subject. This book, a Cambridge Monograph on Mechanics and Applied Mathematics, will be published by the Cambridge University Press this autumn.

DEPARTMENT OF CHEMICAL TECHNOLOGY, UNIVERSITY OF BOMBAY

THE Department of Chemical Technology of the University of Bombay has just celebrated its silver jubilee, and it is thus an opportune time to review briefly the development of this Department and its impact on Indian industries. Its foundation may be largely attributed to the vision and enthusiasm of Sir M. Visvesvaraya, who, as chairman of a Government of India committee, recommended as long ago as 1921 that a faculty of technology be instituted in the University of Bombay and that a College of Technology be established in Bombay City. It is probable that the practical difficulties associated with so large a scheme delayed its fulfilment, for it was not until 1930 that the matter was raised again in a more modest way, with the result that in 1934 the University started two new two-year courses, one in textile chemistry and the other in chemical engineering, to each of which twenty chemistry graduates might be admitted annually. The choice of subjects was a wise one: chemical engineering for its general applicability to all industries, and textile chemistry because of its value to the immense cotton industry of the Bombay Presidency. The first graduates were well received by industry, and the demand soon became far greater than the supply.

Moreover, many private individuals and associations, seeing that this was the beginning of a new era in industrial development, made handsome contributions for the endowment of chairs and readerships, or for the establishment and equipment of specialized laboratories. These have amounted so far to £280,000, and in conjunction with large government grants have made possible the erection on a 14-acre site of a group of laboratories of the most modern type on a unit basis, which allows extension or subdivision with a minimum of disturbance. Special features are the large quantity of small-scale plant and high-class instruments intended mainly for research work. There are a well-equipped workshop and a spacious drawing office in which all students have to work, and the library contains about 8,000 modern technical works and more than 200 periodicals to which readers have free access.

The two sections with which the Department started were originally housed in part of the Royal Institute of Science, which gave little room for expansion; but as soon as the success of the project was assured, preparations were made to build the new laboratories and to add more sections to the Department. Starting in 1943 when the laboratories were ready, new branches were gradually added, and there are now seven two-year courses of study open to chemistry graduates which lead to the degree of B.Sc. (Tech.). These are textile chemistry, and the technology of pharmaceuticals and fine chemicals, food, intermediates and dyes, oils, fats and waxes, plastics, and pigments, paints and varnishes. Chemical engineering is now a four-year course, starting from intermediate, for which the degree of B.Chem.E. is awarded, and there is also a three-year degree course from intermediate in pharmacy. In addition, emphasis is laid on research and work for higher degrees, and there are now 125 postgraduate students out of a total of about 525. Members of the Department have published in all more than 700 scientific notes, articles and papers, mostly on experimental work conducted in the laboratories. The Department also acts as a test house for the textile industry and undertakes sampling, analysis, the investigation of technical difficulties and more comprehensive schemes of industrial research for outside bodies.

The number of students has increased in a remarkable manner. Starting with 109 admitted during the first five years, it rose to 380 ten years later, and during the past four years it has been 785. The proportion taking a degree is very high, more than 1,500 of the 1,700 admitted having been successful. These graduates are much sought after; up to the end of 1957, the last year for which detailed figures are available, 762 were engaged in the manufacturing industry out of a total of 1,316.

Unfortunately, this healthy expansion is threatened by circumstances which must already be a source of anxiety to the authorities—the almost world-wide problem of staff. In Bombay the chairs of textile chemistry, food technology and chemical engineering are all vacant, and in the last-mentioned section one readership is vacant and one lecturer is on study leave, leaving one reader and a temporary lecturer to deal with a four-year course to which forty-five or even more students are admitted annually. It is evident that adequate teaching of the wide range of subjects contained in the syllabus cannot be maintained without a large increase of staff. Other sections are suffering in the same way, although to a smaller extent, and the trouble has been mitigated slightly by members of the staff of other institutions, seventeen of whom are generously helping by giving part-time assistance. It would seem that a satisfactory state of affairs will not be reached until the Department's own graduates and those of similar establishments have increased in numbers and experience to such an extent that the small percentage who take up teaching (augmented perhaps by higher salaries) will be sufficient to provide an adequate staff.

The effect of the introduction of 1,500 trained scientists with an acquaintance with industrial conditions and a specialist knowledge of certain technical operations into the country, followed by more than 150 every year, cannot fail to exert a profound influence on the economy of Bombay and indeed of the whole of India. When it is considered that a large proportion of these men have, in addition, a research outlook which may lead to important improvements, it is difficult to form any estimate as to the changes which may result. It suffices to say that these changes can be only for the good and likely to give the utmost satisfaction to all who so wisely gave their time and their money to further the cause of technical education. H. E. WATSON

THE BRITISH COUNCIL

THE annual report of the British Council for 1958–59 includes a brief tribute to the late chairman, Sir David Kelly, the customary appendixes, particulars of university interchanges, courses and study tours, etc., arranged by the Council in the United Kingdom or overseas, exhibitions and publications, analysis of estimates and accounts, and an assessment by the director-general, Sir Paul Sinker, of the main tasks confronting the Council (The British Council. Annual Report 1958–59. Pp. vi+114+9 plates. London: British Council, 1959). Over the past five years, the Council's gross income, derived mainly from public funds, has almost exactly doubled, from £3 million to £6 million. Its work is carried out in about seventy countries, now some four-fifths in the developing areas of the world, especially in the Near and Middle East, South and South-East Asia and Africa, rather more than half of this being devoted to the recently independent countries of the Commonwealth and to Colonial territories, and the fifth, devoted to Europe, includes an important extension to the German Federal Republic.

Sir Paul Sinker emphasizes three tasks as of pre-eminent importance: educational work, especially but not exclusively in the teaching of English; the fostering of personal contacts between British and overseas people, especially between opposite numbers in the educational, scientific, professional and cultural fields; and helping to ensure that English reading matter is as widely available as possible in the countries with which the Council is concerned. In educational work he comments that the recruitment of British teachers for overseas posts has been a main point of growth during the past two years, and the Council has been concerned to ensure good salaries and living conditions, reasonable security and opportunity for obtaining a suitable job on return. All this has meant additional work for the Council's representatives overseas, and since 1957 these posts have been increased by twenty-five of an educational character. The filling of university posts has presented special difficulty. As regards personal contacts, besides some 40,000 overseas students in Britain, the Council looks after, every year, some 5,000 senior visitors, and Sir Paul directs attention to the proportionate increase in the number of visitors for scientific and technical purposes and to the very considerable increase in the Council's responsibilities for administration of the United Kingdom fellowships and similar schemes on behalf of the Government and other authorities. More than 2,000 Fellows and Scholars a year are cared for under various schemes, and the Council is also responsible for organizing a smaller but important outward flow of specialist tours from the United Kingdom. On the printed word, Sir Paul Sinker stresses the key importance of the library in a British Council centre and the advances on a wide front which have been made possible by the Government announcement on June 22, 1959, regarding the export of British books, etc. Finally, he refers briefly to the opportunities which arise as Colonial territories attain independence and also to those offered by educational television.

The general survey of the year notes that the Council's responsibilities are likely to be increased as a result of the Commonwealth Educational Conference in July 1959, as it is probable that the Council will be asked to assist in implementing the main recommendations of the Conference. About half the £1·5 million by which the Council's grants have been increased since 1957 has been used to implement the decisions announced in the first White Paper regarding the teaching of English overseas. Eighty teachers have been recruited for specific posts, mainly in India, Pakistan, South East Asia, the Middle East and Africa, and almost double this number should be recruited in 1959–60 and a further seventy teachers supplied by the provision of financial assistance to schools. Besides 100 scholarships awarded to bring overseas teachers to the United Kingdom for training, a further 68 are being awarded in 1959–60, and the Council has been encouraged by the growing interest of British universities in these problems. Qualified lecturers and tutors are needed, however, at summer schools for teachers of English both at home and abroad, and a further problem is the provision of material for English language teaching.

Agreement was reached with the government authorities of several countries, notably Pakistan, to extend full Council services to their students in Britain, and during the year 7,500 overseas students were met on first arrival. Some 101 specialist tours, advisory visits and delegations, included one to Poland by Prof. J. N. Davidson, the first of several visits of biochemists arranged at the invitation of the Polish Academy of Sciences, and another to India by Sir Harry Platt. Prof. R. M. Kenedi, of the Royal College of Science and Technology, Glasgow, was invited to visit Lima Engineering University to lecture, assist in organizing programmes of studies and advise on purchase of equipment, and a British engineering library has been opened in the Institute of Higher Studies, Buenos Aires. Under the Commonwealth Scheme of university interchange, awards were made to permit ten short visits for consultation and lectures and 47 travel grants were made to university teachers and postgraduate research workers: under the foreign scheme 121 short visits and three longer visits were arranged, besides 57 visits under the younger research workers interchange scheme. Of 5,153 overseas visitors in the United Kingdom under the auspices of the British Council, 996 were in science and technology, 1,255 in education, 1,152 in social science, and 974 in medicine, 739 coming from the Middle East, 562 from South-East Asia and the Far East, 347 from Latin America, 1,232 from Central and East Europe, and 953 from West Europe. Of the 7,487 students met on arrival, 459 were found permanent accommodation in Council residences, 310 in university halls of residence and hostels, and 3,215 in lodgings. Transit accommodation on first arrival was arranged for 1,877 in Council residences and in lodgings for 1,178, while temporary accommodation was arranged for another 1,354 in Council residences and 4,686 in lodgings, and 9,749 students enrolled at Council centres, 2,353 attending vacation courses of 7–15 days, 1,340 week-end courses, and 8,778 study visits to factories, museums, etc.

CYCLICAL ACTIVITY IN ENDOCRINE SYSTEMS

COMPARED with members of most learned bodies, Fellows of the Zoological Society of London have always been well served—more so than many have realized—in regard to the collections, the library, and the gastronomic and other facilities that they are privileged to enjoy. Additionally, in recent months, there has begun a most welcome series of one-day symposia which will in future take the place of a proportion of the ordinary scientific meetings of the Society. These symposia are not too narrowly specialist, and they are open to all zoologists whether Fellows of the Society or not.

The second such symposium, on "Cyclical Activity in Endocrine Systems", was convened by Prof. E. J. W. Barrington, and held on December 8.

The first two papers dealt with the cyclical activity of the fish thyroid. Dr. A. J. Matty (Nottingham) pointed out that although recently a number of reports have indicated a seasonal change in the thyroid, most have been derived from histological data. Only in one species has hormonal iodine been determined. Variation from species to species both in degree and time of thyroid activity made few generalizations possible, although it is possible in some instances to correlate a seasonal thyroid change with locomotory and migratory changes. Some fish appear to have a more active thyroid during spawning, and Dr. Matty suggested that, as the gland does not seem directly to stimulate gametogenesis, the relationship is due to the influence of the thyroid on protein metabolism, for example, on gonad growth or the development of viviparous young. The thyroid in fish has been shown to influence growth, metamorphosis and integument development and, as current researches have demonstrated that nitrogen metabolism is influenced by thyroxine, it was suggested that a principal role of the piscean thyroid is the control of protein and nucleoprotein metabolism.

Dr. D. R. Swift (Windermere) made a general survey of the external factors which could affect the thyroid and its demonstrated cyclical activity. He emphasized that we do not know what the epithelial cell height and uptake and loss of radioiodine mean in terms of actual activity of the gland and that it was difficult therefore to compare the results obtained by workers using different methods. Dr. Swift suggested that the thyroid undergoes two periods of high activity, one period coinciding with the maturation of the gonads, with which it may be in fact concerned. The second period of high activity occurs as part of what is called the basic cycle of thyroid activity in these animals. This activity of the gland appears to be inversely proportional to the water temperature. It was suggested that temperature plays a major part in the control of the thyroid, but that the length of the photoperiod may influence the gland at low temperatures. It was stressed that it is the change in the environment which is important. Obviously, in Nature, the fish never experience a constant one.

Dr. P. G. W. J. van Oordt, a welcome visitor from Utrecht, is primarily interested in the Amphibia. He spoke of the influence of internal and external factors in the regulation of the anuran spermatogenetic cycle. He concluded that spermatogenesis depends on two so-called final factors, first (as would be expected) the gonadotrophic activity of the pituitary, and secondly, the sensitivity of the germinal epithelium to gonadotrophic hormone. Each such factor is influenced by one or more 'intermediate' factors. These include, for example, a gonadotrophin secretion-inducing agent produced by the preoptic nucleus of the hypothalamus. The male sex hormone is also influential. In Amphibia with a potentially continuous spermatogenetic cycle this activity is primarily regulated by the external temperature. This basic factor, together with the internal rhythm, determines the seasonal changes in the spermatogenetic activity of Anura with a discontinuous spermatogenetic cycle *sensu stricto*.

Dr. A. J. Marshall (London) next discussed cyclical phenomena and corresponding behavioural adaptations in relation to the environment in birds. While paying due regard to the importance of day-length changes as a regulatory factor, he remarked that if we viewed the avifauna of the world, light is important only in species for which it is important that it should be important. A whole range of widely unrelated species would be extinguished by natural forces if they were compelled to reproduce at any particular phase of the cycle of the Sun. In perennially lush areas, it is often biologically beneficial—and possible—for species to have more than one breeding season per year. There is a fascinating, as yet unwritten, story of the changes in behaviour that have gone hand in hand with the neural and endocrine changes dictated by the environment and natural selection.

The complexity of endocrine cycles in vertebrates was emphasized by Prof. E. J. W. Barrington, who gave illustrations from the work of his Nottingham group to show the difficulties encountered in attempting to demonstrate a one-to-one relationship between a particular cycle and a particular function. He concluded that an essential part of the study of such cycles must be the means by which they are coordinated and brought into relationship with the external environment. This involves, in particular, a study of the pituitary and its relationships with the central nervous system. The evolution of the trophic control exerted by the adenohypophysis is one aspect of such a study, and he discussed from this point of view the relationship which has recently been claimed to exist between the adenohypophysis and the endostyle in the ammocoete larva. Results obtained at Nottingham suggest that it is by no means clear that such a relationship is demonstrable on the present evidence. Finally, he considered the problem of the relationship between the hypothalamus and adenohypophysis in teleosts and gave evidence suggesting that (contrary to what has previously been supposed) the vascularization of a particular area of the floor of the infundibulum has at least some of the characteristics of a median eminence, and that glial secretory cells are found in the same area. Identifications of such areas in other fish, and a study of any cyclical variations which they may show, should shed much light on the regulation of endocrine cycles in the lower vertebrates.

Dr. G. W. Harris (London), who thoughtfully made it a condition of his appearance that he would not have to write a paper, dealt comprehensively with comparative aspects of the functional relationships of the hypothalamus and adenohypophysis. He paid a graceful tribute to the pioneer work of the late F. H. A. Marshall in the field of reproduction physiology.

The final two papers were concerned with invertebrates. Dr. M. J. Wells (Cambridge) gave an account of some of his work on the optic glands and the ovary of *Octopus* and described a hormonal system which regulates the onset of sexual maturity. In cephalopods, maturation of the gonad depends upon release of a protein secretion from the optic glands. These are in turn governed by an inhibitory nerve supply originating in the highest centres of the central nervous system. The optic glands can be activated (causing precocious sexual maturity) by severance of this nerve supply, or by peripheral blinding. 'Ripening' of the oviducts and oviducal glands is independent of ovarian maturation. The system is single-stage; the state of these secondary structures is not under the control of sex hormones. The brain-optic gland system of cephalopods has obvious analogues in chordates and arthropods; the factors determining the convergent evolution of mechanisms for delaying the onset of sexual maturity were discussed.

The final paper was by Dr. D. B. Carlisle (Plymouth) on moulting cycles in Crustacea. The act of moulting in arthropods, said Dr. Carlisle, must not be considered as an isolated event but as the most spectacular moment in a continuous cycle of metabolic activity. Each moulting cycle is basically like the one before it, and many of the stages have now been shown to be under hormonal control. Secretion of the 'moult inhibiting hormone' by the endocrine complex of the eye-stalk promotes the formation of new tissues and prevents the Y organ from secreting more than 'marginal' amounts of its hormone. Once this eye-stalk hormone ceases to be secreted the Y organ releases a single 'dose' of a moult-promoting hormone. This initiates the preparations for the moult which, once under way, are co-ordinated by a moult-accelerating hormone from the X organ. The amount of water taken in at the moult itself is regulated by a further eye-stalk hormone. Many other hormones act upon particular moults and modify them in some way, acting mainly upon the stage of proecdysis and making the succeeding moult in some way unique: the moult of puberty and the moult at which a female becomes furnished with the brooding characters are examples. In the same way that the originally similar segments of the vertebrate body have been progressively and uniquely modified (in, for example, the head) so the originally similar moults of arthropods have been modified, until we have what Dr. Carlisle called 'the startling difference' between the larval and imaginal moults of Lepidoptera.

Prof. J. Z. Young, who was chairman of the invertebrate session, enlarged upon a statement made by the final speaker concerning the complications of modern endocrinology. Prof. Young said that the presence of a multiplicity of hormones was the natural consequence of the fact that they were sent out, broadcast through the body and each needed to be addressed to a particular target. No such multiplicity of chemical mediators was necessary in the nervous system, where addresses were specified by connexions. The discovery of more and more distinct hormones was a sign of the maturity of the subject. Endocrinologists were learning to understand the code with which their complicated system operated.

The symposium concluded with a general discussion and a summary by Sir Solly Zuckerman. The papers will be published soon as one of the *Symposia of the Zoological Society of London*. A. J. MARSHALL

AERODYNAMIC CAPTURE OF PARTICLES

A SYMPOSIUM on this subject was held during January 13–14 in the laboratories of the British Coal Utilization Research Association at Leatherhead.

Although this might be thought to be a field of very limited appeal, yet, as evidenced by the wide interests of the fifty scientists present, the impingement of small particles carried in a stream of air and falling on to a solid surface placed alongside or in the way of the flow is a problem which concerns a number of industries, as well as having meteorological implications ranging from the icing of aircraft wings and motor-car windscreens to defence against microbiological warfare.

The wide scope of the applications of this study can be seen by looking through the list of titles offered at this symposium. In 'pure' meteorology one studies cloud formation and collision of raindrops, in applied meteorology the wash-out by rain of particulate matter, whether this be the product of industrial air pollution or of an atomic explosion. A number of cognate problems arise in the laying of mine dusts by water sprays. Then we have the impact of water drops or ice crystals on large moving surfaces such as aircraft wings or turbine blades; when both the obstacle and particles are on a smaller scale we have the action of the gas-mask filter (one of the earliest applications).

The coagulation of particles carried by a stream of air or water is, of course, of great interest and involves many applications where solid or liquid particles have to be transported from place to place, for example, in the industrial process of fluidization.

The basic theory of much of this work stems from Albrecht, who worked out in 1931 the proportion of particles carried by a stream of air which would be intercepted by a cylinder placed athwart the stream. The particles tend to follow the stream lines as they approach the cylinder, but their mass makes them continue in a forthright direction. If their resistance in the stream is given by Stokes's law, that is, direct proportionality to velocity and viscosity, their actual paths depend on this balance between inertia and viscosity in a parameter resembling a particle Reynolds number. This 'particle parameter' has been used continually in capture problems since (and deserves the attribution of Albrecht number).

The conference, opened by a short address of welcome from Dr. Idris Jones, director of research of the National Coal Board, was divided into three sessions. The first morning covered fundamental aspects, a number of developments of the Albrecht thesis, using modern mathematical devices and including other types of flow around the obstacle than the potential one (H. Herne). Prof. P. R. Owen had devoted a considerable study to the process by which a turbulent wind-stream deposits particles on the roof of a tunnel, while Dr. D. J. Brown had studied the behaviour of particles in the process of froth flotation. Dr. T. Gillespie, who had come from the United States to attend the symposium, described his interesting series of researches on the role of electric forces in collision processes.

The afternoon session was devoted to practical applications of capture techniques. Accounts were given of the deposition experiments on radioactive particles and gases carried out at the Atomic Energy Research Establishment (A. C. Chamberlain); on particulate filters—one of the earliest applications—at the Chemical Defence Experimental Establishment (R. G. Dorman); on fish-curing smokes at the Herring Industry's Research Station (W. W. Foster) and two applications in the aircraft industry, namely, impingement of water drops on a surface at supersonic speeds (D. C. Jenkins and J. D. Booker) and on turbine blades (D. L. Martlew).

In the final session the papers dealt with collisions of particles with rain-drops, and of drops with each other. The difficult calculations on the relative motions of particles of comparable size—when the flow pattern around each contestant must be known—had been made by Dr. L. M. Hocking; but other papers were primarily experimental. A long and careful series of experiments by W. Walton and A. Woolcock and less-extensive ones by Dr. R. G. Picknett involved the measurement of the pick-up by a single water drop on which small particles impacted. Finally, the effect of the efficiency of the pick-up process when the particles are wettable and can be engulfed or pass completely through the water drop was discussed both theoretically (by C. S. Pemberton) and experimentally (by B. Oakes).

At the conclusion of the third session, Mr. W. Walton (National Coal Board) thanked the staff of British Coal Utilization Research Association for offering the symposium the hospitality of its conference rooms and laboratories, the work in which the visitors were able to inspect before leaving.

Lively discussions followed the reading of the papers. The whole proceedings are to be published by Pergamon Press, Ltd. and should form an interesting volume in a field which has not been heretofore covered by a book.

E. G. RICHARDSON

THE SHUTTLE-BOX BEHAVIOUR OF ALBINO RATS DURING PROLONGED EXPOSURE TO MODERATE-LEVEL RADIATION

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THIS series of investigations was commenced in an effort to test conclusions reached by other investigators[1,2] that radiation is a 'noxious stimulus' capable of resulting in avoidance conditioning. In three independent attempts to reproduce 'spatial conditioned avoidance' at this Laboratory, no success was obtained. In each of these experiments, albino rats were on alternate days exposed to 1 r. per min. of X-radiation while confined in a black compartment. During subsequent test periods, the rats were given free access to black and white shuttle-box compartment. No significant avoidance of the black compartment was observed in any of the three experiments when the behaviour of the experimental subjects was compared with that of control subjects.

Although no 'conditioned avoidance' was observed n the initial experiments, it could not be concluded that rats are unable to perceive radiation. A more sensitive procedure would seem to be one that allows rats to choose between two shuttle-box compartments —one located in a radiation field and the other shielded from radiation by a lead screen. Because rats normally prefer the darker of two compartments, it was considered appropriate to use shuttle-boxes with one dark and one light compartment and to place the darker compartment in the exposed area. Dose-rates of 0, 1, 2 and 4 r. per min. were employed in this experiment. The results clearly indicated that the experimental animals spent less than a normal amount of time in the usually preferred darker compartment. It was concluded that the presence of radiation in a normally preferred surrounding results in reduced preference for that surrounding; however, no conclusion concerning the nature of the mechanism responsible for the change was possible. Did the rats perceive the radiation, or its immediate consequences, and associate it with the black compartment? Did physiological changes resulting from the radiation produce stimuli which simply overshadowed the normal preference for the dark compartment?

A new experiment was designed which was like the preceding one except that it involved three groups: (*a*) 'radiation experimental' with exposure in the darker compartment and no exposure in the lighter compartment; (*b*) 'non-radiation control' with no exposure in either compartment, and (*c*) 'radiation control' with exposure in both the lighter and darker compartments. The brightness contrast between the two compartments was systematically varied in five steps from black-white to black-black. Both male and female rats were used. It was reasoned that if the previously observed change in shuttle-box behaviour had resulted from the rats learning to discriminate between the exposed and non-exposed areas, then the 'radiation experimental' subjects should again evidence this change, but the 'radiation control' groups should behave no differently from the 'non-radiation controls'. On the other hand, if radiation-produced stimuli simply overshadowed the normal brightness preference, then both of the radiation groups should differ from the 'non-radiation controls' but not from each other. The results clearly favoured the latter alternative, and it was concluded that most of the change in preference for the normally preferred environment was the result of a generalized response to altered physiological state and did not result from the perception of radiation and the learning of an avoidance response. This type of change was significantly more pronounced among female rats. Although the results clearly indicated that the preponderance of the observed effect did not depend upon a perceptual-learning mechanism, the probable presence of such a mechanism was suggested by the results. The two rats in the 'radiation experimental' group which were placed in shuttle-boxes with two otherwise identical black compartments spent an average of only 19 per cent of the time in the exposed compartment. Using a pooled estimate of error derived from the complete results, this value was significantly below chance.

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An experiment was designed using only shuttle-boxes with two identical black compartments. Both male and female rats were used and, in an effort to identify the mechanism responsible for the previously noted sex differences, both male and female gonadectomized rats were also included. For half the rats the left-hand compartment was shielded, and for half the right-hand compartment was shielded. The response of normal animals to the two shield positions was significantly different from that of the gonadectomized animals, that is, on the average the normal animals moved in the direction of the shielded compartment while the gonadectomized rats indicated a slight trend in the opposite direction. Actually, all the observed shift among normal animals was attributable to the normal male group, with normal females indicating virtually no change.

A replication of the black-black shuttle-box experiment with 20 additional normal male rats yielded mean differences in the direction of the shielded compartment almost identical with those for norma males in the preceding experiment; however, large variance resulted in no statistically significant result in this later experiment.

A final experiment to be mentioned involved shuttle-boxes with one black and one medium-grey compartment. For half the rats the black side was shielded and for half the grey side was shielded A statistically significant shift in the direction of the shielded compartment was observed; and the magnitude of the observed shift was greater for male rats, although the sex differences were not statistically significant.

The results of these later experiments in which the only difference between the two treatments is a difference in the location of a protective lead shiel strongly suggest that a perceptual mechanism may take a minor but real role in the behavioural respons of albino rats to low-level radiation and that thi perceptual response is stronger in male rats. Thi latter conclusion is in contrast to the stronger "non-perceptual" response among female rats which wa noted in earlier experiments, and which was incidentally verified in the last mentioned experiment throug comparison with a non-radiation control group.

Radiation exposure produces changes in the amoun of time that an albino rat will spend in a normally preferred surrounding. Two mechanisms are suggested. The major determinant of the change is a generalized response to a radiation-induced physiological state. A perceptual-learning mechanism may take a minor role in the behavioural change observe in male rats.

The dose-rates employed in all experiments excep the one where additional dose-rates are specifically stated was 1 r. per min. in the air inside the shuttle-box compartments.

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[1] Garcia, J., Kimeldorf, D. J., and Koelling, R. A., *Science*, 122, 1 (1955).

[2] Garcia, J., Kimeldorf, D. J., and Hunt, B. A., *Brit. J. Radiol.*, 818 (1957).

ENERGY-LINKED REDUCTION OF MITOCHONDRIAL PYRIDINE NUCLEOTIDE

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IN thermodynamic computations of reactions in biological systems[1] it has been customary to assume that the formation of reduced pyridine nucleotide proceeds in an exergonic reaction in which the free energy of the oxidation-reduction couple $(AH_2 \dashrightarrow A)$ is sufficiently negative with respect to reduced pyridine nucleotide itself that no extern energy source is required to drive the reaction[1]. Th article presents experimental results suggesting th the reduction of mitochondrial pyridine nucleoti can be accomplished by oxidation-reduction coupl having potentials several hundred millivolts high than that of the pyridine nucleotide couple[1–

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This discovery opens up new possibilities for utilization of substrates of high oxidation-reduction potential for pyridine nucleotide reduction with subsequent oxidation in various energy-yielding or synthetic reactions. It also indicates that the respiratory chain can utilize energy conserved at some sites to drive energy-requiring reactions at others.

The remarkable effect of succinate upon the reduction of intramitochondrial pyridine nucleotide was observed early in studies of rat liver mitochondria[4-6] and has recently been studied by Klingenberg *et al.*[18]. This reaction differs sharply from the fumarate oxidation of reduced pyridine nucleotide in non-phosphorylating Keilin and Hartree particles[7]:

$$\text{Fumarate} + \text{DPNH} + \text{H}^+ = \text{DPN}^+ + \text{succinate} \quad (1)$$

The reverse of this reaction requires an energy expenditure since the reductant couple ($A'\text{H}_2 \dashrightarrow A'$) affords insufficient free energy to reduce pyridine nucleotide:

$$\text{DPN}^+ + A'\text{H}_2 + X \sim I = \text{DPNH} + \text{H}^+ + A' + X + I \quad (2)$$

The driving force for such a reaction could be a high-energy intermediate of oxidative phosphorylation ($X \sim I$), which provides the energy required by the difference of oxidation-reduction potentials between the $A'\text{H}_2 \dashrightarrow A'$ couple and the pyridine nucleotide couple. In effect, $X \sim I$ raises the potential of pyridine nucleotide or lowers that of $A'\text{H}_2$ sufficiently to allow the reaction to proceed to completion[8]. This article illustrates the kinetics of reduction of pyridine nucleotide by succinate, particularly in kidney and heart mitochondria, where the extent of reduction of pyridine nucleotide by succinate greatly exceeds that of the usual diphosphopyridine nucleotide-linked substrates. The nature of the substance reduced is identified by spectroscopic and fluorometric data. In addition, the pathway of electron transfer is shown to involve carriers of the respiratory chain, the whole cytochrome chain for oxidation, and part of the chain for reduction. The great sensitivity of the reaction to energy expenditure (for example, by adenosine diphosphate) or to uncoupling of the phosphorylation clearly demonstrates the energy requirements for this reaction. In addition, a direct demonstration of a metabolic recovery process following energy expenditure in pyridine nucleotide reduction has been made.

Material and Methods

Rat liver[9] and guinea pig kidney mitochondria[10], and rat heart sarcosomes[11,12] were prepared by methods described elsewhere. The spectrophotometric[13,14] and polarographic[15] apparatus have been described previously.

Fluorometric measurement of the extent of reduction of mitochondrial pyridine nucleotide[16,17] is used to considerable advantage in rat heart preparations where this component is more difficult to observe than in rat liver preparations. It should be noted that Klingenberg *et al.*[18] (see particularly their Fig. 3*b*) have confirmed the difficulty of making measurements of reduced pyridine nucleotide in rat heart preparations due to the change in absorbancy at 320 mμ[12]; the fluorometric method has much to recommend it for assays of reduced pyridine nucleotide where partial depletion of mitochondrial pyridine nucleotide has occurred and where the chemical method may fail to demonstrate its presence[18].

Succinate-linked Reduction of Pyridine Nucleotide

The data of Figs. 1 *A* and *B* provide examples of spectrophotometric and fluorometric recordings of pyridine nucleotide reduction in kidney and heart mitochondria. The recordings of Fig. 1*A* were obtained with the double-beam spectrophotometer set at measuring wave-lengths appropriate for the reduction of pyridine nucleotide (340 or 350 mμ) and at a nearby reference wave-length (374 mμ). The same phenomenon can be recorded fluorometrically with 366 mμ excitation and 443 mμ measurement[17]. Treatment of the mitochondria in state 1 with adenosine diphosphate gives state 2, a more highly oxidized state of pyridine nucleotide. Addition of 4 m*M* glutamate causes pyridine nucleotide reduction (downward deflexion of the traces) and initiates respiration until the added adenosine diphosphate is exhausted, when a further reduction of pyridine nucleotide is observed (state 4_g). The process may be repeated with a second addition of adenosine diphosphate, as in Fig. 1*B*. In both cases addition of 4 m*M* succinate causes a large, rapid downward deflexion of the trace which these studies indicate to be due to an increased reduction of pyridine nucleotide (state 4_s). Addition of adenosine diphosphate to the mitochondria treated with glutamate and succinate causes an oxidation of the reduced pyridine nucleotide (state 3_s). These experiments demonstrate that the extent of reduction of pyridine nucleotide by succinate considerably exceeds that obtainable with glutamate alone. Mitochondria isolated from ascites tumour cells also show this effect[19]. Bücher and Klingenberg have demonstrated the reduction of pyridine nucleotide in locust sarcosomes by α-glycerophosphate[20,21]. More recently, Klingenberg *et al.*[18] have confirmed our studies on heart and kidney mitochondria. Other pyridine nucleotide-linked substrates of the citric acid cycle have no greater effect than glutamate, and the succinate effect is

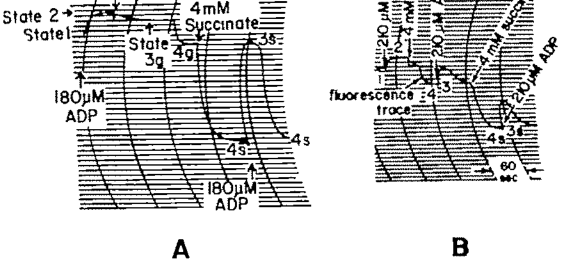

Fig. 1. Comparison of the effects of glutamate and succinate on pyridine nucleotide reduction in guinea pig kidney (*A*) and rat heart mitochondria (*B*). The time-scale increases from left to right; the reagents and states of the mitochondria are indicated in the figures. *A*, spectrophotometric recording at 350–374 mμ; *B*, fluorometric recording, 366 mμ excitation, 443 mμ measurement. Mitochondria suspended in isotonic medium, 26°. (Expts. 658a–10, 791–1)

sensitive to malonate (50 per cent inhibition for malonate/succinate = 0·35).

The pathway of the succinate-linked pyridine nucleotide reduction is not the main pathway of electron transfer; the maximal rate of reduction is about equal to the state-4 rate of oxygen utilization or roughly one-fifth the state-3 rate. Nevertheless, this pathway appears to have considerable theoretical interest and may be of physiological importance.

It is important to clarify the results obtained previously with rat liver mitochondria[22]. In this material, succinate causes reduction of pyridine nucleotide, but its effect is not appreciably in excess of that caused by β-hydroxybutyrate. In more recent comparisons of the effects of glutamate and succinate on rat liver mitochondria, similar to those of Fig. 1, the ratio of the increment on adding succinate (4_s–4_g) to the effect of glutamate alone (4_g–2) is only 0·22. The corresponding average values for heart and kidney mitochondria are 2·1 and 3·3 respectively. The discovery of a large amount of triphosphopyridine nucleotide in mitochondria[20,23] affords an explanation for this difference: apparently triphosphopyridine nucleotide is affected similarly by succinate and β-hydroxybutyrate and thereby obscures accurate spectrophotometric detection of differences in the effect of these two substrates upon the diphosphopyridine nucleotide of liver. For this reason, kidney and heart mitochondria are more suitable for spectrophotometric and fluorometric studies than are liver mitochondria. In previous studies we noted a much smaller pyridine nucleotide–cytochrome *a* ratio in rat heart than in rat liver mitochondria when using α-oxoglutarate as substrate[12]. The larger pyridine nucleotide reduction obtainable with succinate now gives a ratio of ~ 9 : 1, which is about half that for liver mitochondria[24].

Nature of Reduction Product

Spectrophotometric and fluorometric data. To identify the material reduced by succinate and oxidized by adenosine diphosphate, as indicated in Fig. 1, we have recorded spectra representing absorbancy differences between state 4_s and state 3_s in succinate-reduced mitochondria. Fig. 2 also represents studies of two preparations, one from guinea pig kidney (*A*) and one from rat heart (*B*). The increased absorbancy on adding succinate to substrate-free mitochondria (state 2) to give 340 mμ absorption (state 4) is indicated in Fig. 2*A*. Fig. 2*B* shows that the addition of adenosine diphosphate to succinate-treated mitochondria (state 4) causes a band with a maximum at 340 mμ to disappear (note the negative values of the ordinate). The peaks of the spectra are close to 340 mμ, as would be expected for the oxidation and reduction of pyridine nucleotide. Other studies show the succinate-reduced material to have a fluorescence maximum at 443 mμ[16].

It appears that the succinate-reduced material has the physical properties characteristic of the reduced state of pyridine nucleotide. Since, as indicated below, the material so reduced can be oxidized in an amytal-sensitive reaction by the respiratory chain, it is concluded that a reduced form of pyridine nucleotide is formed in this reaction. It is further concluded that the reduced form is bound to some ligand, for example, a protein, because the fluorescence maximum is at 443 mμ[25,26]. Thus this material is denoted DPNH ~ *I*[27] although Slater currently uses the description DPN ~ *I*H (cf. refs. 29, 35). Actually, the two forms could resonate. There are no data available to indicate the chemical nature of ~ *I*. We found that DPNH ~ *I* was refractory to acetoacetate[30,31] or to ferricyanide[32] oxidation, and Purvis believes that it is refractory to low concentrations of alcohol dehydrogenase[33] (but compare below and ref. 34). The interesting possibility that ubiquinone–coenzyme *Q* may be the ligand[35] remains an unsupported hypothesis and now appears to be inconsistent with the reduced pyridine nucleotide and ubiquinone contents of guinea pig kidney mitochondria, which are in the ratio of about 4 : 1 (ref. 36).

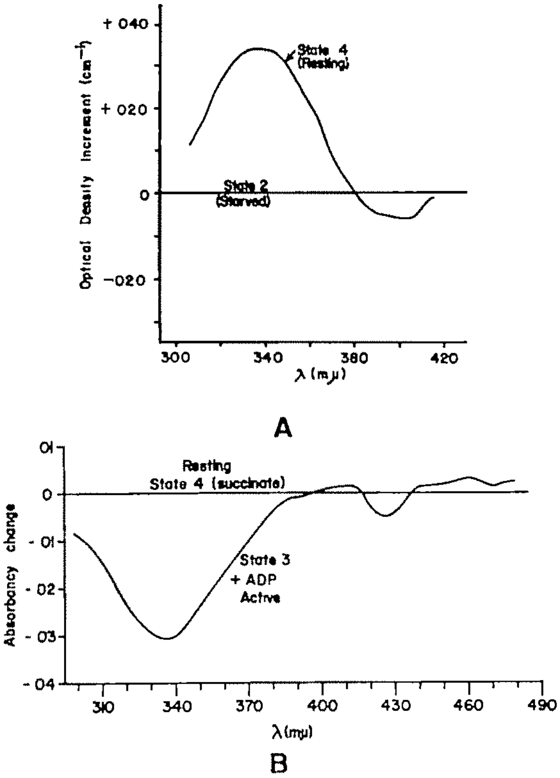

Fig. 2. Spectroscopic changes caused by succinate addition to 'starved' guinea pig kidney mitochondria (*A*) and adenosine diphosphate addition to succinate-treated rat heart mitochondria (*B*). Conditions similar to Fig. 1. (Expts. 664*b*–1, 771*c*–3)

Chemical data. Chemical determinations of the material reduced by succinate, particularly in the kidney mitochondria, are in agreement with the conclusion above[18,37] (also Bücher and Klingenberg, personal communication). The increased absorbancy obtained on adding succinate to the kidney mitochondria is very nearly equal to the decrease of absorbancy caused by adding alcohol dehydrogenase and acetaldehyde to a neutralized sodium carbonate extract of such mitochondria. Thus, it can be said that nearly the same amount of material reduced by succinate in the guinea pig kidney mitochondria can be re-oxidized by dilute yeast alcohol dehydrogenase and acetaldehyde in an extract. This result differs from that obtained by Purvis[38] and Slater[26], who used rat liver mitochondria. It also indicates that diphosphopyridine nucleotide is involved, in agreement with Bücher and Klingenberg[18] (and personal communication). Thus, succinate acts as a powerful reductant of mitochondrial diphosphopyridine nucleotide.

Pathway of Reduction and Oxidation

It is now appropriate to consider the pathway for reduction and oxidation of mitochondrial pyridine nucleotide by succinate. The fact that the oxidation-reduction of succinate-reduced diphosphopyridine nucleotide closely follows the increases and decreases of respiratory activity accompanying the state 4–3–4 transitions suggests that this component is closely associated with the respiratory chain. The oxidation of reduced diphosphopyridine nucleotide has been found to be sensitive to cyanide, antimycin-*A*, and amytal. It can therefore be concluded that pyridine nucleotide reduced by succinate is oxidized by the same pathway as the pyridine nucleotide reduced by the usual diphosphopyridine nucleotide-linked substrates.

Pyridine nucleotide reduction could be activated by a decrease in the velocity of its oxidation or by an increase in the velocity of its reduction, either along the usual pathway or by the activation of a new pathway. We have carefully examined the possibility that the addition of succinate decreases the flow of oxidizing equivalents to reduced pyridine nucleotide by reducing the steady-state oxidation-level of cytochrome *b*. In this case, addition of excess cyanide should cause pyridine nucleotide reduction at a rate exceeding that caused by addition of succinate. This has not been observed in freshly prepared mitochondria with either cyanide or hydrosulphide as inhibitor. In mitochondria depleted of endogenous substrate, addition of cyanide after glutamate causes a slow and incomplete reduction of pyridine nucleotide, whereas addition of succinate causes a complete and three times as rapid reduction. These experiments appear to eliminate a decrease of reduced pyridine nucleotide oxidase activity as a complete explanation. Thus we have considered the succinate effect to be an activation of pyridine nucleotide reduction. Since the increased rate of pyridine nucleotide reduction by succinate is slowed by amytal (see below), we can eliminate an increase of glutamic dehydrogenase activity. The succinate-linked effect probably results from an activation of pyridine nucleotide reduction through an amytal-sensitive flavoprotein interaction, essentially a reversal of electron transfer between cytochrome *b* and reduced pyridine nucleotide via flavoprotein (cf. ref. 18). The reducing equivalents may flow from succinate directly or from glutamate via a small amount of directly reduced pyridine nucleotide[39] to the larger pool of pyridine nucleotide. The above-mentioned results with cyanide and the unpublished results of Klingenberg and Bucher (cf. ref. 18) on α-glycerophosphate support the former view which is embodied in the equations which follow. Ubiquinone–coenzyme *Q* is not yet included in these equations, but experiments on non-phosphorylating preparations would place it next to flavoprotein[40], in which case the conclusions reached here would be unchanged since direct transfer through ubiquinone, as through flavoprotein, would be insensitive to antimycin *A*.

The pathway for the reduction of the material is also indicated by its inhibitor sensitivity. The malonate and amytal sensitivity of the pathway of reduction of pyridine nucleotide which is demonstrated by spectroscopic experiments suggests a transfer pathway similar to that of equation 3, where interaction between flavoproteins (fp_1 and fp_3) causes the reduction of pyridine nucleotide:

$$\begin{tikzcd}
 & & & c_1 & & & \\
 & & & {} & \text{antimycin-}A \arrow[l] & & \\
 & & & b \arrow[uu] & & & \\
 & \text{malonate} \arrow[d] & & & & \text{amytal} \arrow[d] & \\
\text{succinate} \arrow[rr] & {} & fp_1 \arrow[rr] \arrow[uur] & & fp_3 \arrow[uul] & {} & \text{DPN} \arrow[ll, leftrightarrow]
\end{tikzcd} \qquad (3)$$

Such a pathway is in accord with the suggestions of Beinert[41], who favours the idea of direct transfer between flavoproteins. The reduction of diphosphopyridine nucleotide by a diaphorase preparation has recently been studied by Massey[42] and by Searls and Sanadi[43]. However, the tenfold increase in the speed of reduction of cytochrome *b* in the amytal-treated material, together with the fact that the reduction of pyridine nucleotide is slower in the presence of antimycin-*A* than in the presence of cyanide, suggests that reducing equivalents for diphosphopyridine nucleotide pass through the respiratory chain at the level of cytochrome *b*:

$$\begin{tikzcd}
 & & & c_1 & & & & \\
 & & & {} & \text{antimycin-}A \arrow[l] \arrow[d] & & & \\
\text{succinate} \arrow[rr] & {} & fp_1 \arrow[r] & b \arrow[uu] \arrow[rr] & {} & fp_3 & {} & \text{DPN} \arrow[ll, leftrightarrow] \\
 & \text{malonate} \arrow[u] & & & & & \text{amytal} \arrow[u] &
\end{tikzcd} \qquad (4)$$

Since the site of action of antimycin-*A* is between cytochromes *b* and c_1, it is suggested that an inhibition of the ability of cytochrome *b* to transfer reducing equivalents along the chain has occurred. In the case of pyridine nucleotide reduction, antimycin-*A* would be inhibiting the ability of cytochrome *b* to transfer reducing equivalents to the flavoprotein, fp_3, involved in this reduction reaction (see equation 4). The pathways indicated by this equation are in satisfactory accord with the kinetic data on the effect of the indicated inhibitors upon the kinetics of cytochrome *b*, flavoprotein, and pyridine nucleotide, and represent a working hypothesis for discussions of this reaction.

These pathways of equation 4 are written in the usual abbreviated symbolism for the respiratory chain in equation 5:

$$\begin{tikzcd}
 & & \text{succinate} \arrow[r] & fp_1 \arrow[dr] & & & & & & \\
\text{glutamate} \arrow[r] & \text{DH} \arrow[r] & \text{DPNH} \arrow[r] & fp_2 \arrow[r] & b \arrow[r] & c_1 \arrow[r] & c \arrow[r] & a \arrow[r] & a_3 \arrow[r] & O_2 \\
 & & \text{DPNH} \arrow[r, leftrightarrow] & fp_3 \arrow[ur, leftrightarrow] & & & & & &
\end{tikzcd} \qquad (5)$$

This scheme indicates that the pathway for the oxidation of succinate-reduced diphosphopyridine nucleotide proceeds by the same pathway as the reduction. However, this reduced diphosphopyridine nucleotide could equally well diffuse to another set of respiratory carriers and be oxidized there.

Comparison of Succinate and Glutamate Reduction

The chief result of the experiments so far is to show that a larger reduction of diphosphopyridine nucleotide occurs by a succinate-linked pathway through the respiratory carriers than can be achieved by diphosphopyridine nucleotide-linked dehydrogenases. A simple kinetic explanation based upon a greater speed of reduction by succinate appears to be inadequate: the glutamate reduction of diphosphopyridine nucleotide in state 3, as indicated by the respiratory activity, exceeds the rate at which succinate can activate pyridine nucleotide reduction, as illustrated in Fig. 1; fp_2 is rapidly reduced but cannot transfer its electrons rapidly to fp_3 and pyridine nucleotide without the mediation of succinate. Thus an adequate kinetic explanation depends upon a greater rate of electron transfer through fp_2 in the presence of succinate than in the presence of glutamate alone. Also, explanation for the partial reduction of the pyridine nucleotide linked to fp_2 by glutamic dehydrogenase activity is required.

An explanation suggesting that diphosphopyridine nucleotide is reduced by the diphosphopyridine nucleotide-linked dehydrogenases of the mitochondria and triphosphopyridine nucleotide by the succinate-linked pathway has no foundation in the chemical analyses of the kidney mitochondria, which show the presence of very little triphosphopyridine nucleotide[18].

A hypothesis based on chemical difference between the glutamate- and succinate-reduced diphosphopyridine nucleotide in mitochondria which are linked to their reductase system is indicated in the equations below:

$$\text{Glutamate} \dashrightarrow \text{DH} \dashrightarrow \text{DPNH} \sim I \quad (6)$$

$$\text{Succinate} \dashrightarrow (\text{Carriers}) \dashrightarrow fp_3 \dashrightarrow \text{DPNH} \sim I' \quad (7)$$

We here distinguish between the kinds of pyridine nucleotide by the nature of the ligand which inhibits electron transfer in state 4, that appropriate to the glutamic dehydrogenase pyridine nucleotide being designated I and that appropriate to the succinate-linked component being designated as I'. Thus, glutamate has direct access to DPNI and can reduce it directly. Succinate, acting through the respiratory carriers, activates DPNI' through its specific flavoprotein (fp_3). This mechanism explains the way in which succinate and glutamate can reduce different amounts of pyridine nucleotide on the basis of chemical specificity. There are two observations that require an elaboration of this hypothesis. In anaerobiosis, the full amount of diphosphopyridine nucleotide is slowly reduced in the absence of added succinate. Also succinate reduces the total diphosphopyridine nucleotide of the kidney mitochondria in the absence of added glutamate. Both these effects could be explained by diphosphopyridine nucleotide-reduced diphosphopyridine nucleotide transhydrogenase activity[44], which is insufficient to be active aerobically but which nevertheless acts on the two forms of diphosphopyridine nucleotide of equation 6, above, in anaerobiosis.

A slow glutamate-linked transhydrogenase activity would afford a better basis for a kinetic explanation of the different degrees of diphosphopyridine nucleotide reduction by glutamate and succinate. Such an activity could well utilize the pathways of equation 5, fp_1 being more effective than fp_2 in the reduction of pyridine nucleotide via fp_3.

An alternative hypothesis is that diphosphopyridine nucleotide is specifically bound to glutamate- and succinate-linked reducing sites. Such a condition would immediately satisfy the larger reduction of pyridine nucleotide by succinate and a smaller reduction by the usual glutamate-linked and other substrates. Evidence for the binding of mitochondrial pyridine nucleotide has already been presented above from fluorometric experiments. This apparently simple hypothesis becomes more complicated when it is considered that all diphosphopyridine nucleotide-linked substrates apparently reduce the same amount of pyridine nucleotide (α-oxoglutarate, glutamate, malate, β-hydroxybutyrate). Thus, the pyridine nucleotide bound at the glutamate site must be readily available to the other diphosphopyridine nucleotide-linked dehydrogenases (but not to the succinate-linked pathway). Preliminary evidence that adenosine diphosphate (and uncoupling agents) may inhibit the binding of pyridine nucleotide to its succinate-linked site is afforded by fluorometric data which show diminished fluorescence of pyridine nucleotide in anaerobiosis where adenosine diphosphate can accumulate.

There is apparently no difficulty in providing adequate binding sites for pyridine nucleotide at the succinate-linked site, since it is not unusual to have four molecules of pyridine nucleotide bound to a single protein[45].

Compartmentation of an identical chemical species of pyridine nucleotide would also explain the experimental data; it is conceivable that the glutamate-linked material is bound to the cristæ and the succinate-linked material to the matrices. It is necessary to assume that such compartments are slowly equilibrated in anaerobiosis, and that the matrix material can reach fp_3 but not glutamic dehydrogenase in aerobiosis.

Of these hypotheses only the simple kinetic explanation appears inconsistent with available experimental data. Those hypotheses involving: (1) a slow glutamate-linked transhydrogenase activity; (2) chemical specificity; (3) bound pyridine nucleotide; and (4) structural compartmentation must remain under active consideration.

Energy Expenditure in Reduction of Diphosphopyridine Nucleotide

The data above suggest that pyridine nucleotide reduction is associated with not only the reducing power of succinate but also the electron transfer from succinate to oxygen. In fact, the maximal rate of succinate-linked pyridine nucleotide reduction is about equal to the rate of electron transfer in state 4. This correlation, combined with the high sensitivity of the reaction to uncoupling agents, has led us to investigate whether there is a demonstrable energy requirement for this reaction. First, we shall consider the thermodynamic properties of such an energy-linked reaction.

Thermodynamic potential. In considering the energy requirements for the succinate-linked reaction, it is highly desirable to indicate the thermodynamic potential against which this reaction can occur. The graph of Fig. 3 indicates a titration of diphosphopyridine nucleotide of kidney mitochondria with succinate in the absence of fumarate and in the presence of 30 mM fumarate. In the absence of added fumarate, 190 μM succinate gives half-maximal reduction. To obtain an estimate of the potential

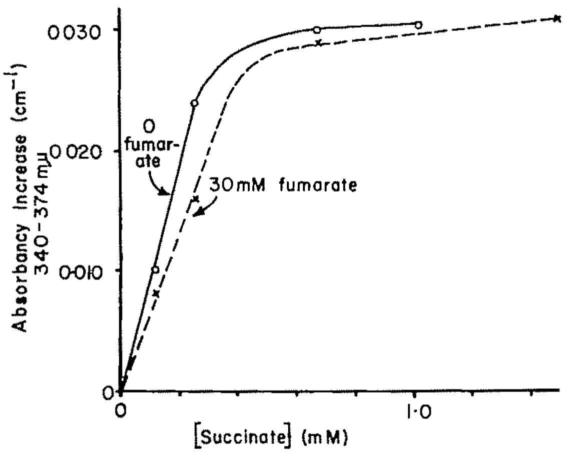

Fig. 3. A titration of pyridine nucleotide of guinea pig kidney mitochondria with succinate in the absence and in the presence of 30 m*M* fumarate. Mitochondria pretreated with 4 m*M* glutamate and then titrated with succinate (cf. Fig. 1). (Expt. 715*d*)

against which this reaction will occur, the titration is repeated in the presence of 30 m*M* fumarate; in this case, 215 μ*M* succinate gives half-maximal reduction. This increase in succinate concentration is so small that the question may be raised as to whether the system is in equilibrium (the fumarate/succinate ratio differs by roughly 10^4 in the two titrations (see also refs. 29, 46)). If we assume an equilibrium, the redox span ranges from 34 mV. (ref. 2) to − 320 mV. (ref. 3) or a total of 354 mV. Even if the assumed equilibrium is proved incorrect, it is clear that the reaction has a considerable energy requirement.

Effect of uncoupling agents. If the overall process involves a high-energy intermediate,

$$\text{Succinate} + \text{DPN}^+ + X \sim I \dashrightarrow \text{fumarate} + \text{DPNH} + X + I + \text{H}^+ \quad (8)$$

uncoupling agents should inhibit the reaction. Table 1 summarizes the concentration of uncoupling agents giving half-maximal effects. It is seen that extremely low concentrations are effective in inhibiting this reaction. More detailed studies identify an inhibition of the reduction as opposed to an activation of the oxidation because of the effects of the uncoupling agent.

It can be seen that the concentration of mitochondrial reduced pyridine nucleotide can be regulated over wide ranges by balancing the reductive effect of succinate against the oxidizing effect of adenosine diphosphate or of substances such as thyroxine which act at such low concentrations that the phosphorylation efficiency would be largely unaffected.

Table 1. CONCENTRATIONS OF UNCOUPLING AGENTS* GIVING 50 PER CENT INHIBITION OF THE RATE OF DPN REDUCTION BY 4 m*M* SUCCINATE (K_m). GUINEA PIG KIDNEY MITOCHONDRIA, SUCROSE-SALT MEDIUM, 26°

Uncoupling agent	DBP	DNP	DIC	DIB	Thyroxine	
Experiment number	722	722	723*b*	723*d*	719*b*	721*b*
Mg^{++}	5 m*M*	5 m*M*	5 m*M*	5 m*M*	5 m*M*	—
Initial state	4 *g*	4 *g*	4 *g*	4 *g*	1	1
Substrate	4 m*M* glutamate	4 m*M* glutamate	4 m*M* glutamate	4 m*M* glutamate	—	—
'K_m' (μ*M*)	22	3 0	1·2	1·0	3 2	0·5

* DBP, Dibromophenol; DNP, Dinitrophenol; DIC, Dicoumarol; DIB, Butyl-3,5-diiodo-4-hydroxybenzoate.

Effect of adenosine triphosphate. In view of the postulated requirement of the succinate-linked reduction for a high-energy intermediate, it appeared reasonable that the addition of adenosine triphosphate would accelerate the reduction reaction. Mitochondria in which the extent of reduction was inhibited ~ 50 per cent by 24 μ*M* dibromophenol were treated with a high concentration of adenosine triphosphate (0·8 m*M*), but no measurable increase of reduction was observed. Apparently the added adenosine triphosphate caused no increase in the concentration of intermediates suitable for reducing the pyridine nucleotide.

Effect of phosphate. In an effort to determine whether the reduction reaction is driven by an $X \sim I$ or an $X \sim P$ compound, the succinate-linked reduction was demonstrated in a phosphate-free medium and found to proceed unchanged in rate or extent. Control experiments based on the lack of a spectroscopic response to the addition of adenosine diphosphate suggested that less than one-twelfth the required amount of $X \sim P$ (see calculations below) could have been present under the experimental conditions.

Recovery from energy expenditure. Equation 8 suggests that the breakdown of $X \sim I$ would result in an activation of oxidative recovery to restore the energy expended in reduction of diphosphopyridine nucleotide. If mitochondria are treated with a low concentration of azide (50 μ*M*), changes in the steady state oxidation-reduction level of the cytochromes due to activation of electron transfer in oxidative recovery are sensitively indicated[47] (Fig. 4). Mitochondria pretreated with glutamate and azide are in state 4. Addition of succinate causes a downward deflexion of the fluorometric trace which corresponds to increasing reduction of pyridine nucleotide. Simultaneously, the trace corresponding to cytochrome *a* shows a cycle of reduction and oxidation which suggests that oxidative recovery is initiated upon adding succinate.

The experiment of Fig. 4 can be made quantitative if a known amount of adenosine diphosphate is added to compare its effect with that of the succinate. Thus, 110 μ*M* adenosine diphosphate is added, giving a larger and longer cycle of reduction of cytochrome *a* than did succinate. (The fluorescence trace shows that, in this instance, reduced pyridine nucleotide

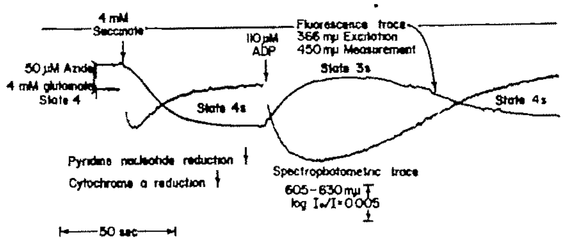

Fig. 4. A demonstration of recovery from energy expended in the reduction of pyridine nucleotide by succinate. Rat liver mitochondria pretreated with 4 m*M* glutamate and 50 μ*M* azide. Energy recovery is indicated by the cycle of reduction and oxidation of cytochrome *a* (measured at 605–630 mμ) caused by the succinate-linked reduction of pyridine nucleotide. 110 μ*M* adenosine diphosphate is added to calibrate the energy recovery (10°). (Expt. 9706–13)

is oxidized, as is the usual case in the state 4-to-3 transition.) A comparison of the areas under the curves corresponding to addition of adenosine diphosphate and succinate reveals that 8 per cent of the area corresponding to 110 μM adenosine diphosphate is caused by succinate addition. In other words, a resynthesis of a high-energy intermediate corresponding to the utilization of 9 μM adenosine diphosphate (22·5 mμmoles) is required. If the amount of mitochondria is varied, a straight-line relationship between the amount of mitochondrial protein and the amount of respiratory resynthesis is found, suggesting that a stoichiometric relationship exists between energy expenditure and pyridine nucleotide reduction. Finally, computing, by suitable calibrations based upon absorbancy changes at 340 mμ, the number of adenosine diphosphate equivalents required to form 1 mole of reduced diphosphopyridine nucleotide, a ratio of $\sim$ 3 : 1 is obtained. It is further found that pretreatment of the mitochondria with amytal does not reduce the energy expenditure upon succinate addition to zero; 30 per cent remains. Thus, for the specific reduction of diphosphopyridine nucleotide by succinate, 2 adenosine diphosphate equivalents are required. We may make preliminary assignments of the energy expenditure at the sites indicated by the following equation:

$$\text{Succinate} \dashrightarrow fp_1 \dashrightarrow b \underset{\overset{\uparrow}{X \sim I}}{\dashrightarrow} fp_3 \underset{\overset{\uparrow}{X \sim I}}{\dashrightarrow} \text{DPNH} \underset{\overset{\uparrow}{X \sim I}}{\dashrightarrow} \text{DPNH} \sim I \qquad (9)$$

This suggests that the succinate-linked reaction not only reduces pyridine nucleotide to the level of − 320 mV. but, further, forms a high-energy bond between it and its ligand, I. However, the technique used in the assay of the adenosine diphosphate equivalents formed is a novel one and some confirmation by independent means would appear to be desirable.

It is not surprising that the energy expenditure in this reduction is high relative to the one adenosine triphosphate equivalent obtained in the oxidation of reduced diphosphopyridine nucleotide because the reduction reaction must pay the full cost of the reaction. For the span between fp_3 and DPNH $\sim I$, the energy conserved as adenosine triphosphate in the oxidation reaction appears to be about one-half of the energy cost of reduction. In this sense, this experiment gives us a rough estimate of the efficiency of oxidative phosphorylation in this part of the chain.

'*Energy transferase*'. It is apparent that high-energy intermediates formed in the cytochrome portion of the chain by succinate oxidation can be used to reduce diphosphopyridine nucleotide. It is also true that glutamate treatment of the mitochondria increases the speed of the succinate-linked reduction, suggesting that the high-energy intermediate formed from diphosphopyridine nucleotide can also participate in diphosphopyridine nucleotide reduction. This is a sort of transhydrogenase activity. In addition, such reduced diphosphopyridine nucleotide can, by its oxidation, form other high-energy intermediates. Thus, there can be an equilibration of high-energy intermediates in the respiratory chain, essentially an 'energy transferase' activity as suggested some time ago by Lipmann[48]. The existence of such a reaction further suggests that intermediates of various energy contents may be used to increase the ratio of reduced to oxidized diphosphopyridine nucleotide until the potential of the system is sufficient to form an intermediate capable of phosphorylating adenosine triphosphate. Such a possibility greatly widens the range of sites that can be considered for energy conservation and may ultimately make the assignment of a particular phosphate bond to a particular redox couple unrealistic. Thus, even more attention may be focused upon the experimentally observed loci, namely, the three sites at which inhibitory interactions are identified by the crossover theorem[49].

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[1] Burton, K., and Krebs, H. A., *Biochem. J.*, **54**, 94 (1953).

[2] Borsook, H., and Schott, H. F., *J. Biol. Chem.*, **92**, 535 (1931).

[3] Burton, K., and Wilson, T. H., *Biochem. J.*, **54**, 86 (1953).

[4] Chance, B., and Williams, G. R., *Fed. Proc.*, **14**, 190 (1955).

[5] Chance, B., and Hollunger, G., *Fed. Proc.*, **16**, 163 (1957).

[6] Chance, B., in "Enzymes: Units of Biological Structure and Function", 447, ed. by Gaebler, O. H. (Academic Press, Inc., New York, 1956).

[7] Slater, E. C., *Biochem. J.*, **46**, 484 (1950).

[8] Glahn, P. E., and Nielsen, S. O., *Nature*, **183**, 1578 (1959).

[9] Lardy, H. A., and Wellman, H., *J. Biol. Chem.*, **195**, 215 (1952).

[10] Hollunger, G., *Acta Pharmacol. et Toxicol., Scand.*, **11**, Suppl. 1 (1955).

[11] Cleland, K. W., and Slater, E. C., *Biochem. J.*, **53**, 547 (1953).

[12] Chance, B., and Baltscheffsky, M., *Biochem. J.*, **63**, 283 (1958).

[13] Chance, B., *Science*, **120**, 767 (1954).

[14] Yang, C. C., and Legallais, V., *Rev. Sci. Instr.*, **25**, 801 (1954).

[15] Chance, B., and Williams, G. R., *J. Biol. Chem.*, **217**, 383 (1955).

[16] Chance, B., and Baltscheffsky, H., *J. Biol. Chem.*, **233**, 736 (1958).

[17] Chance, B., Conrad, H., and Legallais, V., Programme and Abstracts, Biophysical Society, Cambridge, Massachusetts, 44 (1958).

[18] Klingenberg, M., Slenczka, W., and Ritt, E., *Biochem. Z.*, **332**, 47 (1959).

[19] Chance, B., and Hess, B., *J. Biol. Chem.*, **235**, 2413 (1959).

[20] Bucher, Th., and Klingenberg, M., *Angew. Chemie*, **70**, 552 (1958).

[21] Klingenberg, M., and Bucher, Th., *Biochem. Z.*, **331**, 312 (1959).

[22] Chance, B., and Williams, G. R., *J. Biol. Chem.*, **217**, 409 (1955).

[23] Jacobson, K. B., and Kaplan, N. O., *J. Biol. Chem.*, **226**, 603 (1957).

[24] Chance, B., and Williams, G. R., *J. Biol. Chem.*, **217**, 395 (1955).

[25] Boyer, P. D., and Theorell, H., *Acta Chem. Scand.*, **10**, 447 (1956).

[26] Duysens, L. N. M., and Kronenberg, G. H. M., *Biochim. et Biophys. Acta*, **26**, 437 (1957).

[27] Chance, B., Williams, G. R., Holmes, W., and Higgins, J., *J. Biol. Chem.*, **217**, 439 (1955).

[28] Slater, E. C., in Ciba Foundation Symposium on Cell Metabolism, ed. by Wolstenholme, G. E. W., and O'Connor, C. M. (J. and A. Churchill, Ltd., London, 1959).

[29] Slater, E. C., in Proceedings of the Hæmatin Enzyme Symposium, Canberra, Australia, 1959 (in the press).

[30] Chance, B., and Williams, G. R., "Advances in Enzymol.", 17, 65 (1956).

[31] Chance, B., in Ciba Foundation Symposium on Cell Metabolism, ed. by Wolstenholme, G. E. W., and O'Connor, C. M. (J. and A. Churchill, Ltd., London, 1959).

[32] Estabrook, R. W., and Mackler, B., *Fed. Proc.*, **15**, 248 (1956).

[33] Purvis, J. L., Abst., Amer. Chem. Soc., p. 5*C* (Boston, April 1959).

[34] Estabrook, R. W., in Proceedings of the Hæmatin Enzyme Symposium, Canberra, Australia, 1959 (in the press).

[35] Slater, E. C., Spallanzani Lecture, 1959.

[36] Chance, B., *Fed. Proc.* (in the press).

[37] Chance, B., and Hollunger, G. (in preparation).

[38] Purvis, J. L., *Nature*, **182**, 711 (1958).

[39] Devlin, T., *J. Biol. Chem.*, **234**, 962 (1959).

[40] Chance, B., and Redfearn, E. R., Abst., Amer. Chem. Soc., Cleveland (April 1960).

[41] Beinert, H., and Crane, F. L., *J. Biol. Chem.*, **218**, 717 (1956).

[42] Massey, V., *Biochim. et Biophys. Acta*, 30, 205 (1958).

[43] Searls, R. L., and Sanadi, D. R., *Proc. U.S. Nat. Acad. Sci.*, **45**, 697 (1959).

[44] Stein, A. M., Kaplan, N. O., and Ciotti, M. M., *J. Biol. Chem.*, **234**, 979 (1959).

[45] Velick, S. F., in "Mechanism of Enzyme Action", 499, ed. by McElroy, W. D., and Glass, B. (Johns Hopkins Press, Baltimore, 1954)

[46] Chance, B., and Hollunger, G. (in preparation).

[47] Chance, B., and Williams, G. R., *J. Biol. Chem.*, **221**, 477 (1956).

[48] Lipmann, F., in "Currents in Biochemical Research", 137, ed. by Green, D. E. (Interscience Publishers, Inc., New York, 1956).

[49] Chance, B., Holmes, W., Higgins, J., and Connelly, C. M., *Nature*, **182**, 1190 (1958).

LETTERS TO THE EDITORS

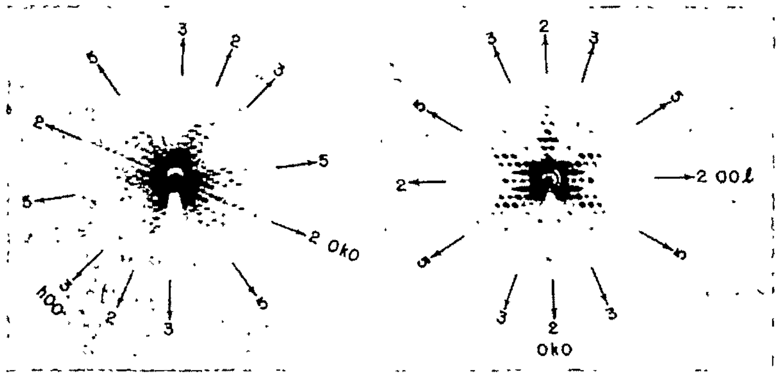
VIROLOGY

Sub-Units in Southern Bean Mosaic Virus

Two results have emerged from preliminary X-ray diffraction studies of single crystals of southern bean mosaic virus. The first, and the more important one, is information about the structure of the virus particle proper, and the second about the packing of the particles in the crystal. Each particle is composed of identical sub-units, and the particles are arranged in hexagonal close-packing to form the crystal. The fact that this virus, like other small 'spherical' ones previously studied, is composed of sub-units again confirms the hypothesis made by Crick and Watson[1]. They suggested that the protein sub-units pack in cubic point-group symmetry and that there are either 12, 24 or 60 such units depending on whether the symmetry of the particle is tetrahedral, octahedral or icosahedral.

Southern bean mosaic virus crystallizes in the orthorhombic system with a probable space group of $C222_1$. There are four particles in the unit cell of dimensions $a = 295$ Å., $b = 508$ Å. and $c = 474$ Å. Because the ratios of cell dimensions are $b/a = \sqrt{3}$ and $c/a = 1{\cdot}61$, and all interparticle distances are probably equal, the virus particles must be hexagonally close-packed in the crystal. Thus, the centre-to-centre distances of the particles in the crystal are 295 Å. This diameter of virus is essentially the same as the value of 286 Å. obtained from small-angle scatterings of X-rays by southern bean mosaic virus particles in suspension[2].

Diffraction patterns of the wet crystals grown in distilled water show reflexions down to spacings of 5 Å., indicating relatively high crystal perfection, with one virus particle very similar if not identical to another. Two of the patterns, $hk0$ and $0kl$, are shown in Figs. 1*a* and *b*. The conspicuous features of these patterns are the regions of high intensity which correspond to the transform of the individual particle. Although the lattice imposes 222 symmetry on the particle, the positions of the high-intensity streaks (rotation axes marked in Fig. 1) are those of the 532 axes of the cubic icosahedral point group. The three perpendicular two-fold axes are parallel to the *a*-, *b*- and *c*-axes of the orthorhombic lattice.



(*a*) (*b*)

Fig. 1. 2° precession photographs of southern bean mosaic virus crystals taken with the X-ray beam parallel to (*a*) the *c*-axis and (*b*) the *a*-axis. The orientations of the five-, three- and two-fold rotation axes of the icosahedral point group 532 are indicated and coincide with the directions of the strongest intensities

If the virus has true icosahedral symmetry, it must be composed of sixty identical or nearly identical protein sub-units. Should the sub-units differ by one or two amino-acids, this would not be detectable in the X-ray patterns. Each sub-unit would have a molecular weight of 85,000, as calculated from the 21 per cent nucleic acid[3] and the molecular weight of the virus of $6{\cdot}63 \times 10^6$ (ref. 4). However, these units may be sub-divided. From amino-acid analysis it is known that the protein chain has a minimum molecular weight of 27,000 (ref. 5). Thus, there are probably three similar protein chains in each of the sixty equivalent units of molecular weight 85,000. The ribonucleic acid, by analogy with the other viruses and from preliminary small-angle scattering of X-rays, is probably located within the protein framework. Little is known about it from the X-ray observations at this stage of investigation, except that if it is not a randomly oriented chain it must have at least four sub-units subject to 222 symmetry. That there may be four sub-units merely means that these units are similar structurally but not chemically.

Southern bean mosaic virus resembles tomato bushy stunt[6], turnip yellow mosaic[7], and polio[8] in that all these particles are probably composed of sixty identical or near-identical protein sub-units which pack in icosahedral symmetry to form the framework of the virus. However, at present, neither the shape of the protein sub-units nor that of the virus itself is known about any of the particles examined. By comparing the data for all these particles, it can be seen that southern bean mosaic virus differs from the other three. The diffraction patterns have a different aspect since the streaks of high intensity are much broader for southern bean mosaic virus. It is not unlikely that the shape of the sub-unit affects the sharpness of the streaks.

Another dissimilarity is in the number of interparticle contact points in the lattice. The other plant viruses crystallize in the cubic system, giving body-centred packing with the contact points of the particles along the three-fold axis of both the cell and the particle. Polio virus, although it crystallizes in the orthorhombic class, packs in a lattice which is scarcely distinguishable from body-centred cubic. In the case of southern bean mosaic virus, due to the hexagonal close packing there are four different contact points: two in the hexagonal plane, one of which is along one of the twofold axes of the cell, and the other along the cell diagonal, close to a five-fold axis of the particle. The contact points of one particle to its three neighbours in the next layer are of two kinds and differ from those in the plane.

More detailed results will be published in the *Contributions from Boyce Thompson Institute*.

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[1] Crick, F. H. C., and Watson, J. D., *Nature*, 177, 473 (1956); "The Nature of Viruses", Ciba Foundation Symposium (J. and A. Churchill, London, 1957).

[2] Leonard, jun., B. R., Anderegg, J. W., Shulman, S., Kaesberg, Paul, and Beeman, W. W., *Biochim. Biophys. Acta*, 12, 499 (1953).

[3] Miller, Gail Lorenz, and Price, W. C., *Arch. Biochem.*, 11, 337 (1946).

[4] Miller, Gail Lorenz, and Price, W. C., *Arch. Biochem.*, 10, 467 (1946).

[5] Magdoff, Beatrice S., Block, Richard J., and Montie, Diane Block, *Contrib. Boyce Thompson Inst.*, 18, 371 (1956).

[6] Caspar, D. L. D., *Nature*, 177, 475 (1956).

[7] Klug, A., Finch, J. T., and Franklin, R. E., *Nature*, 179, 683 (1957); *Biochim. Biophys. Acta*, 25, 242 (1957).

[8] Finch, J. T., and Klug, A., *Nature*, 183, 1709 (1959).

PHYSICS

Pumping Effect of a Gas Discharge High-Vacuum Pump

SIX years ago Foster *et al.*, of the California Radiation Laboratory, constructed a high-vacuum pump with high pumping speed (3,000–7,000 litres/sec.) based on the principle of transport of ions during a discharge in a magnetic field[1]. The limiting pressure reached was less than 1×10^{-8} mm. mercury and the residual gases were free of hydrocarbon vapour. This pump at first caused much interest, but was soon forgotten in favour of more economic pumps based on the pumping effect of evaporated titanium[2].

In our laboratory we have studied some properties of this pump[3]. In the first place, it was necessary to construct a gas discharge pump for experimental work on a small scale, so that it would be relatively easy to make constructional alterations. The discharge tube was made of hard glass (diameter 42 mm., length 1,200 mm.); the inlet opening was at the centre of the tube, the two outlet openings being at the respective ends and connected to a rotary pump by a two-way stop-cock. At both ends an electrode system was point-welded to molybdenum seals. Each electrode system consists of a cathode (a heated tungsten filament and molybdenum target) and a molybdenum ring anode fixed to two long molybdenum tie-beams enclosed in glass. In direct proximity to the anodes in the direction towards the centre, the tube was narrowed for a length of about 5 cm. to a diameter only a little larger than the diameters of the holes in the anodes (28 mm.). The solenoid of the vacuum-pump, which was run by a d.c. power supply (consumption 4 kW.), was placed on the outside of the tube. Parts of the solenoid which were in the neighbourhood of the outlets had more turns of wire than the others; in this way it was possible to obtain a sufficient intensity of the magnetic field throughout the region of the outlet openings, which was necessary for focusing the discharge without the necessity for using an internal solenoid (as in the apparatus of ref. 1). The combination of ring anodes and narrowing of the tube makes it unnecessary to have the anodes very close to the discharge tube.

The second problem was to find the dependence of the pumping speed on pressure by varying the inlet opening (that is, for different diameters of the inlet opening, for different distances between the solenoids and therefore for different intensities of the magnetic field in the region of the inlet opening). The pumping speed was deduced from the pressure drop, which was measured by a cold-cathode ionization gauge and by a McLeod gauge, both of which were connected to the evacuated region. The maximum pumping speed with an inlet opening containing three tubes of total cross-section 1·9 cm.^{2} was 18 litres/sec. (ref. 3). In this case the distance between the solenoids was 12 mm. The maximum pumping speed for one tube of 3·2 cm.^{2} cross-section was 34 litres/sec.; in this case the distance was 32 mm. The mean intensity of the magnetic field was 1,250 Oe., anode voltage 280 V. The dependence of pumping speed on pressure is shown in Fig. 1*a*. The pumping speed reaches a maximum for pressures of the order of 10^{-5} mm. mercury and decreases to the limiting pressure of 8×10^{-7} mm. mercury of this system.

According to present views, there are two processes involved in this pump: the transport of ions from the region of the inlet opening through the anodes into the regions of the cathodes, and sorption processes. Foster *et al.*[1] attributed the pumping effect to the transport of ions. This explanation was criticized and the effect was attributed to sorption processes, since pumping continued even after the fore-vacuum pump was disconnected. In order to show how the two effects participate in the total pumping effect the following experiments were carried out.

It was found that pumping continues (in agreement with Foster *et al.*[1]) even after the disconnexion of the rotary pump. During the measurement of the pressure drop in the evacuated system, which was carried out from the moment of closing the stop-cock connecting the discharge pump with the rotary pump, the pressure drop in the outlet openings was also measured by a gauge. This gauge showed a drop in pressure, which was, however, much smaller than

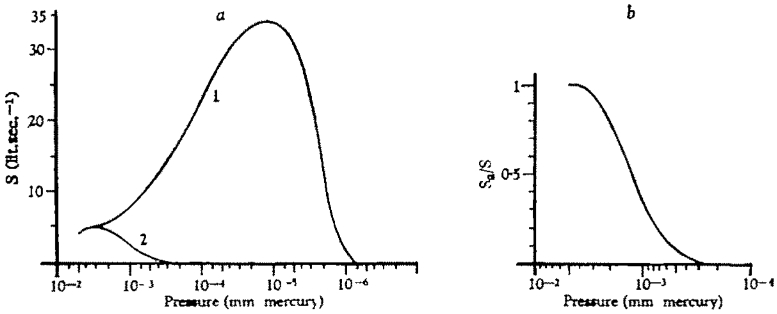

Fig. 1. Course of the pumping speed and the fraction of the total pumping effect allotted to the sorption mechanism in dependence on pressure. (*a*) Curve 1, pumping speed in the evacuated region; curve 2, pumping speed in the fore-vacuum region. (*b*) Dependence of the fraction of the total pumping effect caused by the sorption mechanism on pressure

the corresponding drop in the evacuated region. By comparing the pumping speeds in the evacuated and fore-vacuum region (Fig. 1*a*) it is found that the pumping speed S_a in the fore-vacuum region (which cannot be attributed to the transport process) as a fraction of the total pumping speed S is strongly dependent on pressure (Fig. 1*b*), and for pressures less than $2 \cdot 4 \times 10^{-4}$ mm. mercury the pump evacuates on the principle of transport and recombination of ions. It is plausible that the transport of ions becomes of importance at pressures where the mean free path becomes greater than the linear dimensions of the tube. (The values of pumping speed in the fore-vacuum region were also evaluated from the drop in pressure, that is, assuming constant volume of gas; this supposition is not quite correct, because molecules constantly flow from the high-vacuum region into the fore-vacuum region; that is why the cited values of pumping speed are only approximate.)

Finally, an experiment was carried out in which one of the anodes was inclined from its position. Hence the focused discharge was also inclined, so that a gap arose between the discharge column and the wall of the tube in the narrowed region of the tube connecting the fore-vacuum with the high-vacuum region. In the space of this gap (shaded by the inclined anode from the discharge column) molecules could enter from the fore-vacuum region into the high-vacuum region and could not be ionized on their path. By measurement it was actually found that in this case the value of the pressure in the high-vacuum region was almost equal to the pressure in the outlet openings. This effect could not be attained if the evacuation was based on sorption processes.

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[1] Foster, J. S., Lawrence, E. O., and Lofgren, E. J., *Rev. Sci. Instr.*, 24, 388 (1953).

[2] Herb, R. G., Davis, R. H., Divatia, A. S., and Saxon, D., *Phys. Rev.*, 89, 897 (1953).

[3] Pátý, L., and Neužilová, R., *Czech. J. Phys.*, 8, 746 (1958).

Optical Scanning of Liquid Concentration in Flow Studies with Porous Models

It is often convenient to employ laboratory models for the study of fluid behaviour in oil reservoirs. Typical experiments may concern the flow of two liquids miscible with one another. Basically, this calls for measurements of concentration as a function of time and position. If the liquids can be distinguished by some parameter, a corresponding method of observation may suggest itself. Thus, Beran[1] employed radioactively tagged liquids in his models, while Rifai[2], de Josselin de Jong[3] and others measured the electrical conductivity of salt solutions flowing through packed beds.

Such methods have the advantage of allowing the entire model to be monitored, but they suffer in varying degrees from lack of resolution. The porous medium is generally simulated by a packing of granular material. Retention of a realistic permeability requires that the grains be kept relatively fine; hence the main obstacle to optical scanning resides in the fact that thick packings are normally opaque.

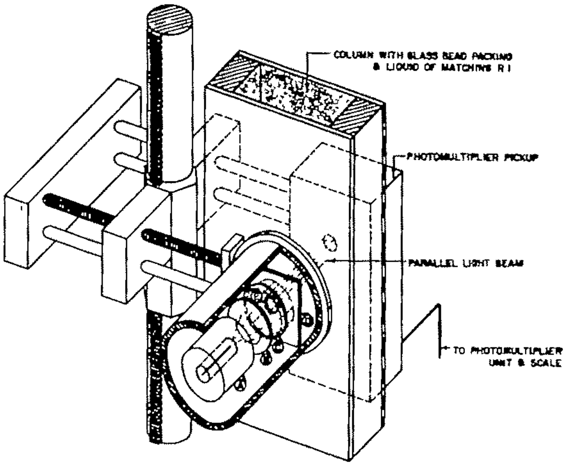

Fig. 1. Scanning unit for dispersion studies. *A*, Point source; *B*, heat filter; *C*, lens; *D*, interference filter; *E*, exit stop

In the method here described, the refractive indices of liquid and packing substance are chosen to be identical at the laboratory temperature, thus enabling a three-dimensional model to transmit light. Optically matched systems of this kind are called Christiansen filters[4,5], and are essentially a suspension of low-dispersion particles, for example, optical glass, in a highly dispersive liquid. Light transmission will be restricted to a narrow range of wave-lengths at the intersection of the two dispersion curves. The system thus acts as a monochromator the region of transmissivity of which will shift if the temperature of the liquid is changed.

In the present arrangement (Fig. 1) the above principle applies with changed emphasis. Thus, the light is rendered monochromatic prior to entry into the flooded model, which is maintained at constant temperature. At the exit face, the pencil passes through a narrow aperture into the photomultiplier unit, which registers constant intensity in the presence of a single liquid phase. When two inter-miscible liquids are under study, one of the components is identified by a dye, thus allowing concentration profiles in the mixing zone to be mapped colorimetrically.

A number of applications of the Christiansen system to model studies have been suggested (Steber and Marsden[6], van Meurs and van der Poel[7], Heller[8]). The last-named employed a matrix of ground crystalline calcium fluoride, which offers the advantages of a low refractive index ($n_D = 1 \cdot 4338$) and optical isotropy. Spherical glass beads have been chosen here as an alternative packing material, being hard, chemically inert and available in bulk, besides offering surfaces which may be readily conditioned from water-wet to oil-wet preference. Anisotropy, however, now arises due to small deviations between spheres, of the order of 0·001 unit from the mean refractive index. In the present model (Table 1), where the search beam axis may cut as many as 300 glass-liquid interfaces, the above variation will suffice to cause scattering and absorption of most of the incident light. This disadvantage has been rendered unimportant by prefiltering, careful collimation and use of a sensitive detector.

The container was assembled from plates of '*CR*-39' ('Homalite') thermosetting plastic. Set up vertically, the column is first flooded with clear

Table 1. PROPERTIES OF MATERIALS USED IN THE MODEL

Glass Bead Packing:
  Diameter of spheres, 0·18 ± 0·08 mm.
  Density, 2·86 gm./cm.^{3}
Liquid Mixture (all data quoted for 25° C.)
  Components—(*a*) chlorobenzene
  (*b*) 1,2,4-trichlorobenzene
  Dye, blue alkyl aminoanthroquinone
  Volume proportions (*a* : *b*), 60·9 : 39·1
  Density of mixture, 1·236 gm./cm.^{3}
  Viscosity of mixture, 0·81 ± 0·01 cp.
  Refractive index of mixture, $n_D^{25} = 1·5408$
  Temperature gradient of R.I., $\Delta n/\Delta T = -0·00050/°$ C.
Column:
  Height, 100–120 cm.
  Internal cross-section, 5·0 cm. × 2·5 cm.

liquid (Table 1). In the simpler initial experiments this is displaced with an identical mixture, which in addition carries oil-soluble dye. Both phases are injected through a two-chamber unit at the base, designed to produce at time zero of the experiment a sharp and level interface between the two liquids.

The light from an argon-sealed arc is rendered yellow by a Bausch and Lomb transmission-type interference filter, with peak at 5890 Å. and half-width 80 Å. The collimated pencil is about 1·8 mm. in diameter, that is, one order of magnitude larger than the glass beads. A much narrower beam would probably register local inhomogeneities in the matrix. Lamp and photomultiplier pick-up move as a unit for scanning along two co-ordinates (Fig. 1), thus allowing transverse as well as longitudinal dispersion to be monitored in models of suitable width.

Finally, the necessary temperature control may be deduced from a curve of relative beam intensity I/I_0 versus temperature T of the clear liquid, with maximum brightness at the matching value (25° C). It is more convenient first to plot the variation of I/I_0 as a function of refractive index, with T constant. This has been done by mismatching clear mixtures in the porous model, and using the temperature gradient, $\Delta n/\Delta T$ (Table 1), to convert the result into the required curve (Fig. 2). In our arrangement, an increase in dye concentration (c) from 50 to 51 per cent will reduce I/I_0 by 1·7 per cent, and this in turn corresponds to a temperature fluctuation of less than 1° C. In a typical mixing zone of 10 cm. length (c = 10–90 per cent), the colour contrast involved should be just observable. Consequently, temperature errors of ±1° C. or more affect the measurements and must be avoided through suitable controls.

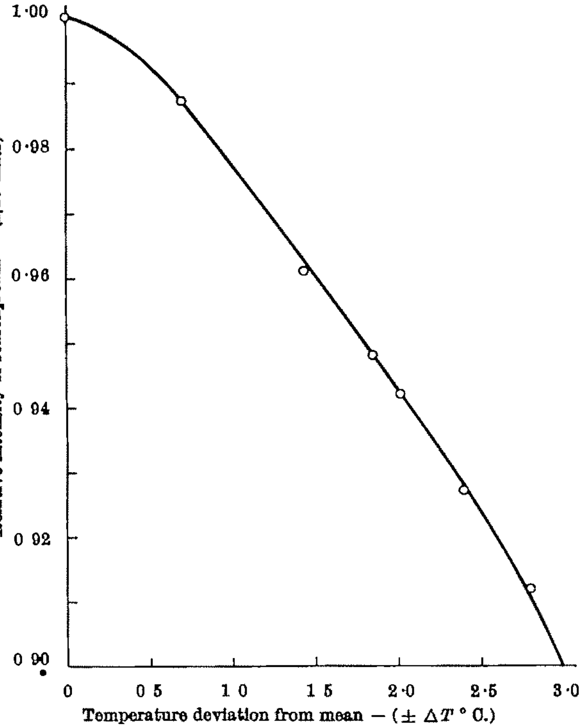

Fig. 2. Reduction in the brightness of the search beam due to temperature deviations from the matching value (for $n_D^{25} = 1·5408$)

The method is adaptable to a variety of interesting problems in reservoir physics; but it will be applied initially to a theoretical study of miscible flow. Solutions to the differential equations of two alternative statistical models are available[9], and the experimental method described should be well suited to furnish the data on which to base a decision.

I am indebted for much valuable advice to Mr. J. W. Young, manager of production research, Imperial Oil, Ltd., Calgary, and Prof. H. E. Gunning of the Chemistry Department, University of Alberta, Edmonton. My thanks are also due to Dr. W. N. McKay, of this Laboratory, for the viscosity determination quoted in Table 1.

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[1] Beran, M. J., Ph.D. Dissertation, Harvard University (1955).

[2] Rifai, M. N. E., Ph.D. Dissertation, University of California (Berkeley, 1956).

[3] de Josselin de Jong, G., *Trans. Amer. Geophys. Un.*, **39**, 1160 (1958).

[4] Christiansen, C., *Ann. Physik und Chemie*, **23**, 298 (1884).

[5] Summer, W., "Photosensitors", 300 (Chapman and Hall, London, 1957).

[6] Steber, J. D., and Marsden, S. S., *Producers Monthly*, **20**, 48 (1956).

[7] van Meurs, P., and van der Poel, C., *J. Petrol. Technol.*, **213**, 103 (1958).

[8] Heller, J. P., *Rev. Sci. Instr.*, **30**, 1056 (1959).

[9] Scheidegger, A. E., *Trans. Amer. Geophys. Un.*, **39**, 929 (1958).

Gallium Arsenide Tunnel Diodes

ESAKI, or tunnel, diodes[1-3] in gallium arsenide have been made in this Laboratory. Zinc-doped gallium arsenide was prepared by diffusion to give a concentration of 9×10^{19} impurity atoms/cm.^{3} and tin pellets were alloyed on to this material to form a junction. The diameter of the pellets varied from 0·003 to 0·015 in.

Diodes made in this manner have shown negative resistances of between 0·1 Ω and 4 Ω, with a wide range of peak currents from 70 m.amp. to 7 amp. The highest ratio of peak to valley currents observed was 30 : 1, and the average value was about 15 : 1. The voltages at which the peak and valley currents occurred were not constant but varied from device to device, from 0·13 to 0·50 V. for the peak current, and from 0·35 to 0·55 V. for the valley current. These variations can be partially explained by the presence of series resistance of about 0·1 Ω, probably in the contacts to the devices.

The diodes were found to switch across the unstable negative resistance region in a time of less than 1 mμsec., this being the limit of the measuring equipment. When the diodes were mounted in a strip-line designed to resonate at a frequency of about 400 Mc./s., relaxation oscillations were observed. The fundamental frequency varied with d.c. bias voltage over the range 300–560 Mc./s. and in all cases harmonics were seen up to 1 kMc./s., which was the upper limit of the receiver. 5 milliwatts output power was obtained.

We wish to thank the Admiralty for permission to publish this communication.

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[1] Esaki, L., *Phys. Rev.*, **109**, 603 (1958).

[2] Sommers, H. S., *Proc. Inst. Rad. Eng.*, **47**, 1201 (1959).

[3] Gremmelmaier, R., and Henkel, H., *Z. Naturforsch.*, **14**, *a*, 1072 (1959)

GEOPHYSICS

Micropulsations in the Earth's Magnetic Field Simultaneous with Pulsating Aurora

Some preliminary results have been obtained from an investigation being undertaken at College, Alaska, concerning the correspondence of the Earth's magnetic field micropulsations[1] with pulsating aurora for the purpose of explaining the physical mechanism giving rise to the phenomena. Fig. 1 illustrates the nightly coincidence of micropulsations, measured with a 6-ft. diameter magnetic north axis loop antenna of 21,586 turns and 3914-Å. auroral pulsations from 70° of sky measured with a lens, interference filter, photo-multiplier system. Fig. 2 shows an occasion of simultaneous oscillations from the two phenomena.

Messrs. A. Belon, C. Deehr, M. Rees and G. Romick, of the Geophysical Institute Optics Division, have aided me in the development of the experiments.

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[1] Campbell, W. H., *J. Geophys. Res.*, **64**, 1819 (1959).

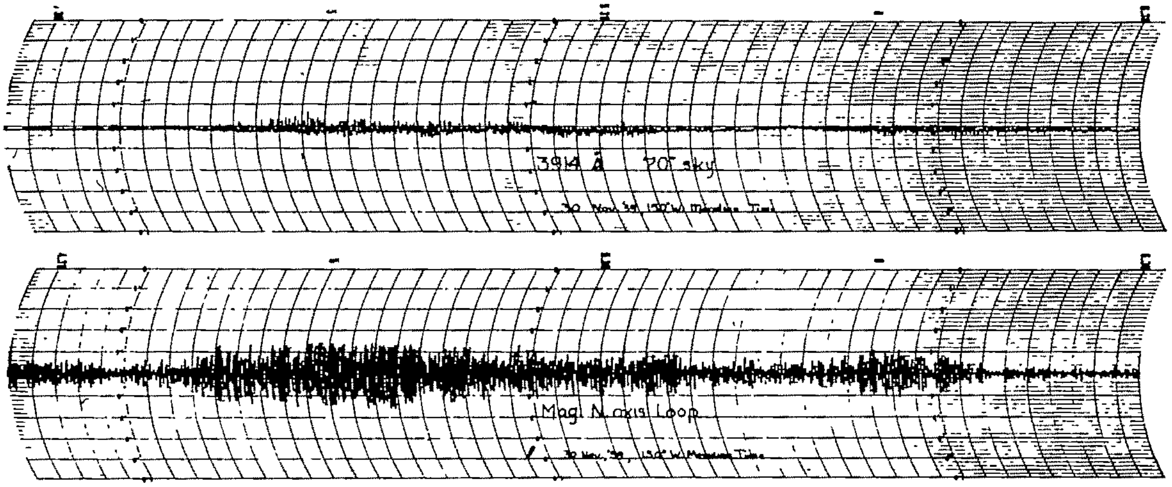

Fig. 1. Coincident pulsating aurora and magnetic micropulsations (12γ full scale)

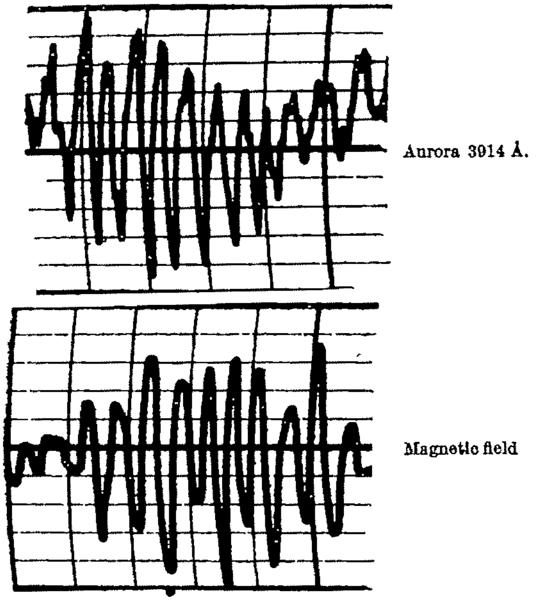

Fig. 2. Simultaneous oscillations of 3914 Å. aurora and micropulsations in the magnetic north–south direction

An Expanding Earth with Loss of Gravitational Potential Energy

Recently, and independently, Carey[1] and Heezen[2] have concluded that the Earth is expanding. The first question asked of such a hypothesis usually takes the form of an inquiry as to the source of energy for such an expansion. This question is based on the assumption that the gravitational potential energy of the Earth is inversely proportional to its radius, and the assumption in turn is presumably based on the well-known equation (see, for example, Loney's "Statics", p. 359).

$$E = -\frac{3}{5}\frac{GM^2}{a}$$

where E is the gravitational potential energy, G the gravitational constant, M the mass and a the radius of the Earth. However, the equation refers only to a sphere of uniform density, and one possibility that appears to have been overlooked is that a non-uniform Earth can expand and lose gravitational potential energy at the same time.

The following expressions for density ρ_r versus radius r in the Earth have been obtained by fitting linear density distributions of the form: $\rho_r = \rho_0(1 - kr)$ to each section of Bullen's[3] model B of the Earth:

$0 < r < 1{\cdot}250 \times 10^8$ cm.	$\rho_r = 17{\cdot}900(1 - 1{\cdot}29162 \times 10^{-9} r)$
$1{\cdot}250 < r < 1{\cdot}389$,,	$\rho_r = 42{\cdot}078(1 - 5{\cdot}14625 \times 10^{-9} r)$
$1{\cdot}389 < r < 3{\cdot}471$,,	$\rho_r = 13{\cdot}408(1 - 7{\cdot}53631 \times 10^{-10} r)$
$3{\cdot}471 < r < 6{\cdot}291$,,	$\rho_r = 7{\cdot}505(1 - 7{\cdot}69678 \times 10^{-10} r)$
$6{\cdot}291 < r < 6{\cdot}338$,,	$\rho_r = \text{constant} = 3{\cdot}34$
$6{\cdot}338 < r < 6{\cdot}371$,,	$\rho_r = \text{constant} = 2{\cdot}79$

If spherical symmetry is assumed these distributions give the right mass, $5 \cdot 977 \times 10^{27}$ gm., and a slightly high moment of inertia, $8 \cdot 120 \times 10^{44}$ gm. cm.^{2}; the discrepancy between this value and the generally accepted one is immaterial for the purposes of the present argument. The gravitational potential energy of an Earth with the above density distributions is $- 2 \cdot 47949 \times 10^{39}$ ergs.

If in the past the density distribution in the Earth was of the form $\rho_r = \rho_0(1 - kr^2)$ from $0 < r < a$, where a is its past radius, then assuming that the mass has remained constant and that the surface density of the Earth was $2 \cdot 85$ gm. c.c.$^{-1}$, one model with a higher potential energy but a smaller radius than the present Earth is $a = 6 \cdot 27 \times 10^8$ cm. and $\rho_r = 10 \cdot 197(1 - 1 \cdot 83275 \times 10^{-18} r^2)$. The potential energy of this model is $-2 \cdot 47464 \times 10^{39}$ ergs and its moment of inertia is $8 \cdot 036 \times 10^{44}$ gm. cm.^{2}. If there are no external or internal sources of energy the Earth will tend to proceed from a higher state of potential energy to a lower state, and one way it could do this would be to expand to its present form. Thus expansions of the order of 100 km. seem quite possible without postulating any source of energy. The Earth will, in fact, lose energy during the expansion.

If it is assumed that the mass of the Earth has remained constant and that $\rho_a = 2 \cdot 85$ gm. c.c.$^{-1}$ then Earth models of smaller radii but higher potential energies can be found for any value of $n \geqslant 1$, where $\rho_r = \rho_0(1 - kr^n)$. The higher the value assumed for n the greater is the possible expansion. Furthermore, for each value of n the range of models with likely density distributions covers models with higher or lower moments of inertia than the present Earth. If density distributions which, on present evidence, seem improbable are postulated, then expansions of the order of 1,000 km. are possible. Expansions are also possible even if the region $0 < r < 3 \cdot 471 \times 10^8$ cm. is kept constant and only the mantle and crust are changed.

In this communication no attempt has been made to postulate possible mechanisms. These, and other possible models, will be discussed in a fuller account elsewhere.

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Jan. 15.

[1] Carey, S. W., "The Tectonic Approach to Continental Drift", Continental Drift Symposium, 177 (University of Tasmania, Hobart, 1958).

[2] Heezen, B. C., "Paleomagnetism, Continental Displacements, and the Origin of Submarine Topography", International Oceanographic Congress Preprints, 26 (Amer. Assoc. Adv. Sci., Washington, 1959).

[3] Bullen, K. E., *Mon. Not. Roy. Astro. Soc., Geophys. Supp.*, 6, 50 (1950).

BIOPHYSICS

Effect of Ultra-violet Light on the Shape and Size of Deoxyribonucleic Acid

SEVERAL investigators[1,2] have found that the viscosity of solutions of deoxyribonucleic acid is reduced by irradiation with ultra-violet light of 254 mμ. This change could be the result either of main-chain scission which reduces the length of the molecules or of an increase in flexibility which allows the molecule to take up a more coiled configuration and thereby reduces the volume occupied in solution, as measured by the radius of gyration.

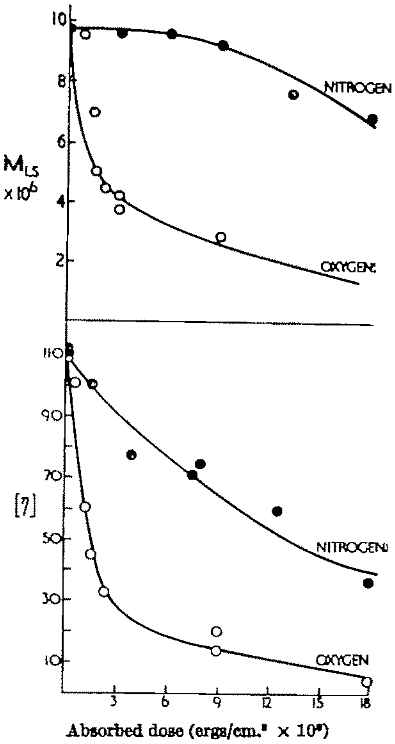

Fig. 1. Changes in the weight average molecular weight (M_{LS}) and in the intrinsic viscosity ([η] in dl./gm.) of deoxyribonucleic acid following irradiation with ultra-violet light of 254 mμ at the rate of 1×10^4 ergs/cm.^{2}/sec. The polymer was at a concentration of 0 01 gm./dl. and either oxygen or purified nitrogen was bubbled through the solution before and during the irradiation

We have carried out simultaneous measurements of the molecular weight by light scattering and of the viscosity in a Couette viscometer on solutions of deoxyribonucleic acid irradiated with a 44-watt 'Hanovia 769/67' mercury resonance lamp in a 'Vycor' envelope. The water-cooled irradiation vessel was specially constructed to allow total immersion of the lamp in the deoxyribonucleic acid solution and gas bubbling through the solution during the irradiation. The deoxyribonucleic acid was obtained from viable salmon sperm and contained less than 0·1 per cent of protein. Different preparations, varying in molecular weight, were used for the experiments shown in Fig. 1 and in Table 1. All measurements were made in 0·2 *M* saline containing citrate buffer.

Fig. 1 shows that, in the absence of oxygen, the first effect of ultra-violet light of 254-mμ wavelength is to reduce the viscosity without altering the molecular weight. As the dose is increased the molecular weight falls, showing that the molecule is severed, but the principal effect remains one of coiling.

If oxygen is present, then the molecular weight falls immediately; but the change in viscosity is very much greater than that due to the reduction in molecular weight alone. The introduction of oxygen gives rise to a second process, main-chain scission, without decreasing the amount of coiling that is produced when the irradiation is done under pure nitrogen.

This oxygen effect is not due to attack by ozone since the 'Vycor' filter effectively excludes the oxygen-

Table 1. Effect of Added Substances on Ultra-violet-induced Changes of Molecular Weight (M_{LS}) and Radius of Gyration (ρ_z in Å.) of Deoxyribonucleic Acid

Dose (ergs/cm.^{2} × 10^6)	No additive		Dimethylaniline 10^{-4} M		Glutathione 10^{-4} M		Glucose 10^{-1} M		Catalase 0·01 per cent	
	M_{LS}	ρ_z	M_{LS}	ρ_z	M_{LS}	ρ_z	M_{LS}	ρ_z	M_{LS}	ρ_z
None	6·7	2,420	7·7	2,600	6·3	2,400	6·3	2,420	7·7	2,400
14 (Nitrogen)	6·0	1,240	5·7	1,370	6·1	1,570	5·5	1,450	7·7	1,750
3·6 (Oxygen)	2·5	1,060	5·7	1,980	5·1	1,610	7·3	2,120	4·5	1,850

absorbing bands of ultra-violet light. Experiments in which oxygen that had been passed over the 'Vycor' lamp and was then bubbled through a solution of deoxyribonucleic acid failed to reveal any degradation.

Coiling of deoxyribonucleic acid is produced when the specific hydrogen bonding between the bases in the twin helix is disrupted[3]. Our results would suggest that this structure is disorganized by ultra-violet light before there is any extensive photochemical degradation of the bases. Internal conversion can lead to the degradation of excitation energy to thermal energy, and it would appear that the increase in the oscillational degrees of freedom brings about this change of shape. Photolysis of the pyrimidine or purine bases would not decrease the molecular size except by initiating further reactions such as hydrolysis.

Direct interaction between an oxygen molecule and the singlet excited state to form peroxides which are further decomposed by the ultra-violet light can best explain the main-chain scission in the presence of oxygen. Photochemically produced OH radicals have been shown[4] to reduce the viscosity of solutions of deoxyribonucleic acid and we were able to show by light-scattering measurements that this effect was due mainly to main-chain scission.

Dimethylaniline, glutathione and glucose were all found to protect the molecular weight and radius of gyration of the irradiated deoxyribonucleic acid against the oxygen effect (see Table 1), presumably by competitively reacting with the degradative free radicals produced via the peroxide. Protection by catalase was observed and is probably due to the decomposition of the peroxide in such a way that it does not give rise to degradative free radicals. Poisoning of the catalase by potassium cyanide caused a loss in protective activity. In the absence of oxygen none of these substances reduced either the coiling or the main-chain scission process significantly, except, oddly, catalase.

A puzzling feature is that the biological effects produced by ultra-violet light have not been found to be influenced by oxygen. Possibly cell constituents act as protective agents, and from this it would follow that cellular lesions in so far as they are due to attack on deoxyribonucleic acid follow from an interference with the hydrogen bonding between the bases and not from a degradative reaction. Doses needed to produce detectable coiling are very large when compared with those necessary to produce changes in cells. Probably the number of hydrogen bonds that have to be broken to produce sufficient coiling to be measured by physicochemical techniques is very much greater than that necessary for interfering with the biological function of deoxyribonucleic acid.

This work has been supported by grants to the Chester Beatty Research Institute (Institute of Cancer Research: Royal Cancer Hospital) from the British Empire Cancer Campaign, the Jane Coffin Childs Memorial Fund for Medical Research, the Anna Fuller Fund and the National Cancer Institute of the National Institutes of Health, U.S. Public Health Service.

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[1] Hollaender, A., and Oliphant, J., *J. Bact.*, **48**, 447 (1944).

[2] Errera, M., *Biochim. Biophys. Acta*, **8**, 30 (1952).

[3] Thomas, C. A., and Doty, P., *J. Amer. Chem. Soc.*, **78**, 1854 (1956).

[4] Butler, J. A. V., and Conway, B. E., *Proc. Roy. Soc.*, B, **141**, 562 (1953).

Blood Flow through Narrow Tubes

In the course of an extensive investigation (with Prof. A. C. Burton at the University of Western Ontario) of the hæmodynamics of perfused vascular beds, it was found necessary to examine the somewhat simpler case of blood flow in narrow glass tubes. Accordingly, the pressure-flow curves in tubes of radii 50–750μ for human erythrocytes suspended in standard acid–citrate–dextrose were measured at 25·5° C. over a wide range of hæmatocrits[1–4]. A typical set of such pressure-flow curves is shown in Fig. 1. The curves were found to approach linearity at moderate flow rates; if the linear segments are extrapolated back, they intersect the positive pressure axis at finite values and then converge to a common 'nodal point' on the negative flow axis. However, in the non-linear region at very low flow rates, the curves appear to converge toward the origin. There is no evidence of any yield stress or intercept on the positive pressure axis. All the curves can be represented by an equation of the form:

$$\dot{Q} = MP - B[1 - \exp(-kP)] \qquad (1)$$

where $\dot{Q}$ is the volume rate of flow and P the pressure gradient; the parameters M and k depend on both tube radius and hæmatocrit, whereas the nodal point B depends only on the tube radius[3]. Pressure-flow curves of similar shape have been obtained by other workers and can be inferred from the apparent viscosity data of Kümin[5].

The results obtained from the basic pressure-flow measurements were used to plot the rate of shear at the wall versus the shear stress (consistency curves). It can be shown[2] that the shear-rate is given by:

$$F = \frac{1}{\pi R^3}\left[3\dot{Q} + P\frac{d\dot{Q}}{dP}\right] \qquad (2)$$

$$\text{or:} \quad F = \frac{4\dot{Q}}{\pi R^3} + \frac{B}{\pi R^3}\left[1 - (1 + kP)\exp(-kP)\right] \qquad (3)$$

and can be plotted from (3) as a function of the shear stress $PR/2$. The first term in (3) is the shear-rate

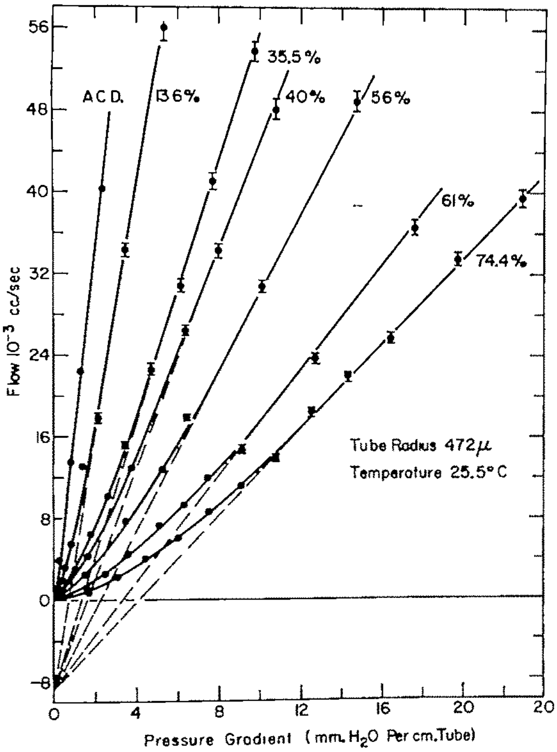

Fig. 1. Pressure-flow curves for human red cells suspended in acid–citrate–dextrose for various hæmatocrits as indicated in volume per cent

for a Newtonian liquid while the second term arises from the non-linearity of the pressure-flow curves. A set of such consistency curves for three hæmatocrits is shown in Fig. 2. They have approximately the same shape as the pressure-flow curves, and become linear at shear stresses greater than about 20 dynes/cm. From the data of Kumin[5], it is possible to plot what might be called the 'apparent shear rate' as a function of shear stress as shown in Fig. 3. This is a plot of the usual consistency variables (for a fluid that obeys Poiseuille's law), so that the contribution of the second term in equation 3 is not present. The error introduced by this approximation is less than 10 per cent, and it does not affect the basic curve shape. The important features of all these curves are the non-linearity below about 20 dynes/cm., and the absence of any finite intercept on the stress axis.

Equation 1 can be given a phenomenological interpretation by assuming that the fractional change in differential fluidity is proportional to the change in shear stress[4]. Such a fluidity change can be interpreted physically as a structural re-organization of the fluid with increasing flow-rates that arises from the axial accumulation of the red cells in a variable shear field[2].

The Fahraeus–Lindqvist effect appeared very prominently in the data of Kümin[5] and also in our own experiments[3,4]. The tube radius at which the apparent viscosity is reduced 10 per cent below the asymptotic value for large tubes was found to vary from 130μ at 10 per cent hæmatocrit to 633μ at 80 per cent. If the effect is interpreted as a sigma phenomenon[6], the average thickness of the unsheared laminæ was found to vary from 3·5μ at 10 per cent hæmatocrit to 34μ at 80 per cent. If, on the other hand, the effect arises from the presence of a cell-free marginal zone[7,8], it was found that its thickness must be 6μ at 10 per cent hæmatocrit, 3μ at 40 per cent, and only 1·5μ at 80 per cent. Such a cell-free marginal zone could also result from the axial accumulation of the red cells. It is difficult to determine the relative importance of the marginal zone and the

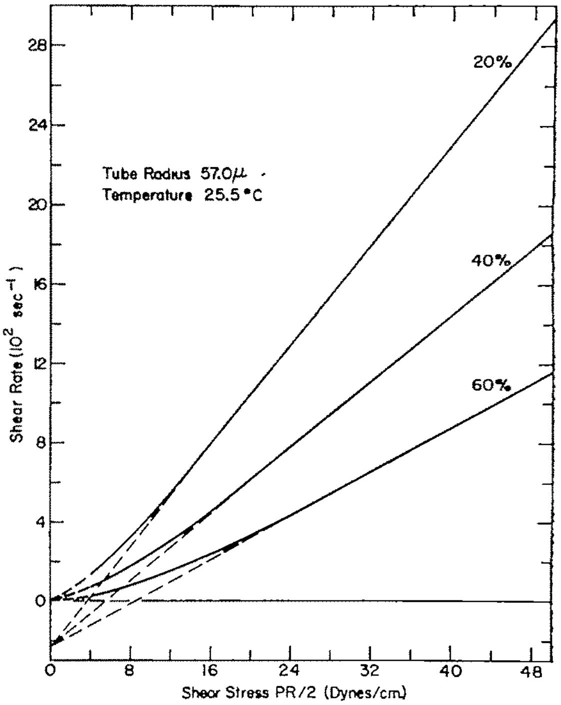

Fig. 2. Consistency curves for human red cell suspensions of three hæmatocrits. The broken curves near the origin represent the only extrapolation beyond the measurements

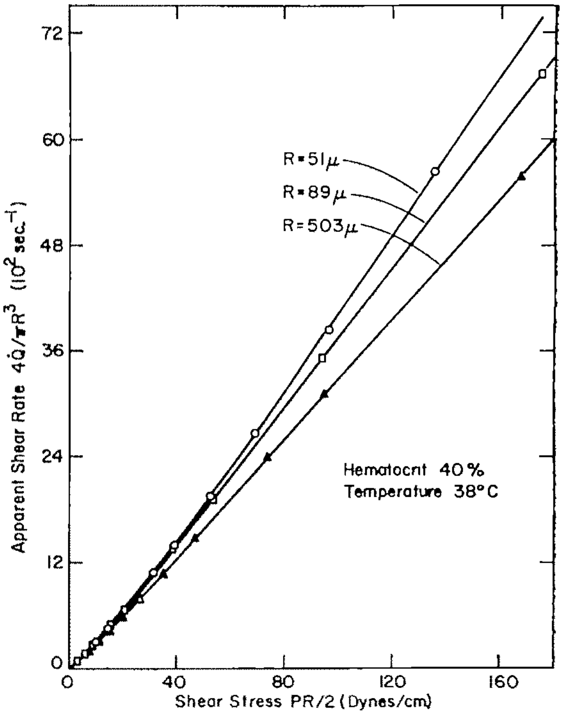

Fig. 3. Consistency curves for defibrinated ox blood calculated from data of Kumin

sigma phenomenon in producing the Fahraeus–Lindqvist effect.

On the basis of some measurements of A. L. Copley, Scott Blair[9] and Reiner and Scott Blair[10] have recently suggested that the rheological properties of blood can be described in terms of a theory put forward by Casson[11] for varnish and ink. This theory is applicable to substances the consistency curves of which intersect the stress axis at a yield value. However, the consistency curves which we have obtained for blood do not seem to intersect the stress axis at a finite value, but rather become non-linear below 20 dynes/cm. and point toward the origin. The lowest shear stress that was reached in Copley's measurements was 100 dynes/cm., and so the non-linearity of the consistency curves was not detected. Thus it does not appear that Casson's theory would provide a satisfactory description of the flow properties of blood at very low shear stresses if no yield value exists.

I wish to thank Dr. M. G. Taylor, now at the University of Sydney, for several helpful discussions, and Mr. J. E. Baptist, of this laboratory, for his help with the calculations.

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[1] Haynes, R. H., *Canad. Biol.*, 15, 255 (1956).

[2] Haynes, R. H., and Burton, A. C., "Proceedings of the First National Biophysics Conference", ed. by Quastler, H., and Morowitz, H. J., 452 (Yale University Press, 1959).

[3] Haynes, R. H., and Burton, A. C., *Amer. J. Physiol.*, 197 (5), 943 (1959).

[4] Haynes, R. H., "The Rheology of Blood", doctoral dissertation (University of Western Ontario, London, Canada, 1957).

[5] Kumin, K., Inaugural Dissertation, Universität Bern (Paulusdruckerei, Freiburg in der Schweiz, 1949).

[6] Dix, F. J., and Scott Blair, G. W., *J. App. Phys.*, 11, 574 (1950).

[7] Taylor, M. G., *Phys. Biol. Med.*, 3, 273 (1959).

[8] Taylor, M. G., *Aust. J. Exp. Biol. Med. Sci.*, 33, 1 (1955).

[9] Scott Blair, G. W., *Nature*, 183, 613 (1959).

[10] Reiner, M., and Scott Blair, G. W., *Nature*, 184, 354 (1959).

[11] Casson, N., "The Rheology of Disperse Systems", ed. by Mill, C. C., Chap. 5 (Pergamon Press, London, 1959).

CHEMISTRY

Crystal Spectrum of Copper Biguanides

IN a previous communication[1] it was shown that the absorption spectrum of a microscopic crystal of copper-ethylene-diamine-*bis*(acetylacetone) consists of three bands as predicted by the ligand field theory. Copper biguanides form transparent reddish crystals[2], very convenient for measurement in our experimental arrangement. In Fig. 1 are given the spectrophotometric optical density (*O.D.*) vs. wave-length (λ) curves for a thin crystal of copper *bis*-biguanide dibromide dihydrate with light polarized along the long axis of the crystal and at right angles to it, respectively. It is evident that the first curve consists of three bands, but the sharpness and fine structure are not as well defined as was the case with copper-ethylene-diamine-*bis*(acetylacetone). The decomposition of the curve into its component bands shows three peaks at 485 mμ, 540–550 mμ and 600 mμ. When the light was polarized at right angles to the long axis of the crystal, the absorption spectrum was found to consist of two bands, with peaks at 485 mμ and 550 mμ, the peak at 600 mμ being missing. If we associate the band of longest wave-length with the transition $d_{z^2} \rightarrow d_{xy}$, then the result may mean that the *z*-axis of the molecular field coincides with the long axis of the crystal.

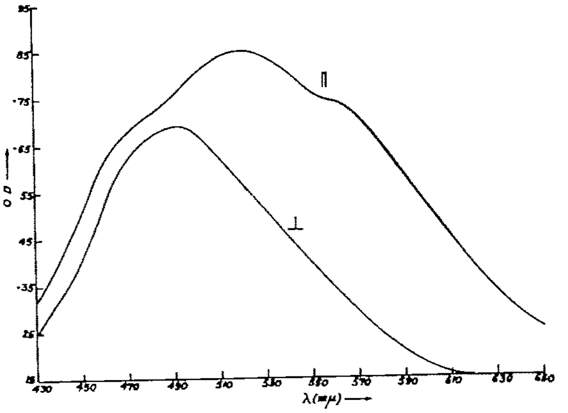

Fig. 1

The crystal spectrum of the chloride and also of the fluoride was found to be identical with that of the bromide, indicating that the anion was outside the co-ordinating sphere and had no perturbing action on the copper *d*-orbitals, which is also corroborated by the chemical properties[2]. In solution, all these compounds have one broad absorption band and show no structure resembling a crystal spectrum. Further, the structure could be detected with extremely thin crystals only. As the thickness of the crystal was increased, the structure of the band became increasingly less well defined.

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[2] Ray and Bagchi, *J. Ind. Chem. Soc.*, 16, 617 (1939).

Structure of Lithium Methoxide

RECENTLY, Bradley has proposed a theory which correlates and predicts the structures of metallic alkoxides[1]. At the moment, Bradley's theory cannot be fully developed because of the lack of direct structural information. In particular, it is not known whether lithium methoxide is ionic, as generally assumed, or whether it is a giant polymer[2]. This communication describes the structure of lithium methoxide, which is shown to be similar to that of lithium hydroxide[3]. Although the structure has some features reminiscent of a giant two-dimensional polymer, it would normally be considered ionic.

Lithium methoxide was prepared by solution of the metal in methanol. Excess methanol was pumped off, and specimens of the resultant powder transferred to thin-walled 'Pyrex' capillaries which were sealed with 'Picien' wax. All operations were carried out in a dry box. Powder photographs were taken with a 3-cm. 'Unicam' oscillation camera. The photographs showed that the unit cell was tetragonal with $a = 3{\cdot}55_5$ and $c = 7{\cdot}68_7$ Å. Systematic absences corresponded to the space group *P4/nmm* (No. 129). The density calculated on the assumption of two formula units in the cell was 1·30 gm./c.c. The observed density, which could not be accurately determined because the powder was so hygroscopic, lay between 1·2 and 1·5 gm./c.c. The lithium ions lie at $\pm(\frac{1}{4}, \frac{3}{4}, 0)$, the oxygen atoms at $\pm(\frac{1}{4}, \frac{1}{4}, u_1)$, and the carbon atoms at $\pm(\frac{1}{4}, \frac{1}{4}, u_2)$. The methoxide ion has four-fold symmetry so that the hydrogen atoms, which were ignored in the analysis, presumably adopt a statistical arrangement to conform with this symmetry. The parameters u_1 and u_2 were determined by trial and error and found to be $0{\cdot}10_5$ and 0·29 respectively. Thus the O—CH_3 distance is 1·42 Å.

If the structure is viewed in a direction normal to the four-fold axis, it is seen to consist of layers *ABCBAABCBA* . . . , with $A = CH_3$, $B = O$ and $C = Li$. The methyl groups are separated by 4 09 Å. in the [*c*] direction and by only 3·55 Å. in the [*a*] and [*b*] directions. The oxygen atoms are bonded to a methyl group on one side and are co-ordinated to four lithium ions on the other to form square pyramids with oxygen atoms at the centre. The Li . . O distance is 1·95 Å. and the LiOLi angle 80·3°. The lithium ions are surrounded by a tetrahedron, flattened in the [*c*] direction, of oxygen atoms. The OLiO angles are 131·1° and 101·7°. The Li . . Li distance is 2·51 Å. and the nearest O . . O contact 2·99 Å. The $LiOCH_3$ angle is 114·5°.

A full account of this work and of further physicochemical measurements on this and other alkali metal methoxides will be published elsewhere.

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[1] Bradley, D. C., *Nature*, 182, 1211 (1958).

[2] Bradley, D. C., and Faktor, M. M., *Nature*, 184, 55 (1959).

[3] Ernst, Th., *Z. phys. Chem.*, B, 20, 65 (1933). Dachs, H., *Z. Krist.*, 112, 60 (1959).

New Univalent Cobalt Complexes of the $[Co^{I}(CN)_5X]$ Type

Hume *et al.*[1] found that the aquopentacyanocobaltate(III) and cobaltate(II) complexes were irreversibly reduced in one step to the cobalt(I) state polarographically in the cyanide medium, and the cobalt(I) complex was neither reducible nor oxidizable at the dropping mercury electrode.

Recently, the similar cobaltate(III) complexes of pentacyano type, $K_{2+n}[Co^{III}(CN)_5X]$, were also found to be reduced irreversibly in one or two steps to the univalent cobalt complexes at the dropping mercury electrode[2], where X denotes the ligand, Cl^-, Br^-, I^-, SCN^-, N_3^-, $S_2O_3^{2-}$, NO_2^-, or SO_3^{2-}.

In order to clarify the structure and the co-ordination number of this univalent cobalt complex, a number of attempts have been made to isolate the complex from the solution, but have failed mainly due to the great solubility of the complex in alcohol and water.

From the following two reasons, however, it was concluded that the univalent cobalt complex has a similar structure to the original oxidized form, that is, the hexa-co-ordinated formula, $[Co^{I}(CN)_5X]$.

(1) It was ascertained in advance polarographically that the yellow solution of the univalent cobalt complex was easily obtained from the reduction of the pentacyanocobaltate(III) ion by 3 per cent sodium amalgam in an atmosphere of nitrogen. No deposition of metallic cobalt was found in any case. From this solution of the cobaltate(I) complex, the pentacyanocobaltate(II) complex, $K_3[Co^{II}(CN)_5]\cdot H_2O$ (ref. 3), was always isolated in an atmosphere of nitrogen as bright violet crystals by addition of ethanol in portions with respect to all the pentacyanocobaltate(III) complexes. This fact means that the pentacyano structure was not destroyed and was still maintained in the univalent cobalt complex.

(2) Generally, all these cobaltate(I) complexes were oxidized by air to the cobalt(III) state on exposure. By pursuing the increase of the wave-height with time, however, it was found that there existed considerable differences among the rates of oxidization, depending upon the kind of ligand, X. For example, the cobaltate(I) complex obtained from the reduction of azido- or nitro-pentacyanocobaltate(III) ion was seen to be instantly oxidized to the cobalt(III) state, on being exposed to the air. On the other hand, the oxidation-rate of the cobaltate(I) complex obtained from the other pentacyanocobaltate(III) ions was found to be small, though the cobaltate(I) complex was still oxidized slowly in open air. This fact implies that the sixth ligand, X, is still co-ordinated about the central cobalt(I) atom in the univalent cobalt complex.

The re-oxidized product obtained from these univalent cobalt complexes was also identified as a hydroxypentacyanocobaltate(III) complex, $K_3[Co^{III}(CN)_5OH]$ (yellow crystals). This is due to the alkalinity which resulted from the sodium amalgam used. Therefore, the bond between Co^{I} and the sixth ligand, X, is considered not to be so strong. Hence, in a solid state, it might be possible that the penta-co-ordinated complex, $K_4[Co^{I}(CN)_5]$, can exist rather than the hexa-co-ordinated complex, $K_4[Co^{I}(CN)_5OH_2]$, being considered analogous to the case of the penta-*iso*nitrilo complexes[4], $[Co^{I}(NCR)_5]Y$ (yellow crystals), where R indicates phenyl-, *p*-tolyl-, or *o*-tolyl-radical and Y, the ion, ClO_4^-, I^-, NO_3^-, or ClO_3^-.

Accordingly, the processes of the electrode reaction for the pentacyanocobaltate(III) complexes were considered to be as follows. For the halogeno complexes:

$$
\begin{array}{llll}
[Co^{III}(CN)_5X]^{3-} + e^- & \longrightarrow & [Co^{II}(CN)_5X]^{4-} & \text{1st step} \\
[Co^{II}(CN)_5X]^{4-} + e^- & \longrightarrow & [Co^{I}(CN)_5X]^{5-} & \text{2nd step} \\
[Co^{I}(CN)_5X]^{5-} + H_2O & \longrightarrow & [Co^{I}(CN)_5OH_2]^{4-} + X^- & \\
 & & X = Cl^-, Br^-, I^- &
\end{array}
$$

For the thiocyanato complex (in the pentacyano complex, $K_3[Co(CN)_5SCN]$, the ligand, SCN^-, is bound to the central cobalt(III) atom by sulphur, while in the ammine complexes, for example $[Co(NH_3)_5NCS]Cl_2$, the ligand, NCS^-, is co-ordinated by nitrogen atom. Therefore, in this case, the equation of the complex can scarcely be considered):

$$
\begin{array}{llll}
[Co^{III}(CN)_5SCN]^{3-} + e^- & \longrightarrow & [Co^{II}(CN)_5SCN]^{4-} & \text{1st step} \\
[Co^{II}(CN)_5SCN]^{4-} + e^- & \longrightarrow & [Co^{I}(CN)_5SCN]^{5-} & \text{2nd step}
\end{array}
$$

For the azido-, nitro-, thiosulphato- and sulphito-complexes:

$$
[Co^{III}(CN)_5X]^{(2+n)-} + 2e^- \longrightarrow [Co^{I}(CN)_5X]^{(4+n)-} \quad \text{1st step}
$$

$$
X = N_3^-, NO_2^-, S_2O_3^{2-}, SO_3^{2-}
$$

The resulting pentacyanocobaltate(I) complexes did not give the anodic wave at all. Namely, all the above electrode reactions are irreversible polarographically, though the reversibility of the reduction waves looks fairly large in appearance.

The mechanism of the electron transfer and the electronic configuration for the above reactions are believed to be as follows. The acceptance of the electrons occurs in two stages:

$$d^2d^2d^2[D^2SP^3] \xrightarrow{\bar{e}} d^2d^2d^2[D^2SP^3]d^1 \longrightarrow$$

$$d^2d^2d^2d^1[DSP^2PD] \xrightarrow{\bar{e}} d^2d^2d^2d^2[DSP^2PD]$$

First, the electron is accepted into the 4*d* peripheral orbitals temporarily, where it is held loosely, since the 3*d* atomic orbitals of Co(III) atom are fully occupied. After that the consecutive change of the electronic configuration and the placement of the electron in the stable 3*d* orbitals occur slowly. Therefore, this electrode process requires fairly high overpotential. In the second stage, the electron is accepted into the vacant 3*d* orbital directly. Thus, the difference of the electronic configuration in the oxidized and the reduced forms is considered to be one of the main causes for irreversibility of the electrode reactions.

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[1] Hume, D. N., and Kolthoff, I. M., *J. Amer. Chem. Soc.*, 71, 867 (1949).

[2] Maki, N., Fujita, J., and Tsuchida, R., *Nature*, 183, 458 (1959).

[3] Adamson, A. W., *J. Amer. Chem. Soc.*, 73, 5710 (1951).

[4] Malatesta, L., and Sacco, A., *Z anorg. allg. Chem.*, 273, 247 (1953)

Carbonates of Uranium

MILLER and co-workers[1] for the first time isolated uranyl carbonate, prepared by the action of carbon dioxide under pressure on uranium trioxide, ammonium and sodium uranates. A different process was followed by Chernyaev and co-workers[2], who prepared the compound by the treatment of aqueous and alcoholic solutions of uranyl nitrate with carbon dioxide under pressure or aqueous suspensions of uranium oxide, UO_2, under normal conditions. A simple and convenient method has been developed by us for the preparation of the compound. The hitherto unreported compound uranium (IV) oxycarbonate, the preparation of which is described below, on exposure to dry air undergoes simultaneous oxidation and dehydration, resulting in the formation of the compound UO_2CO_3, 2·5 H_2O, the progress of the oxidation being indicated by the gradual change of colour from dark green to bright yellow. This hydrated uranyl carbonate, when heated *in vacuo* at 160°, gives the anhydrous compound UO_2CO_3, which is stable up to 500° C.

Uranyl ion undergoes photoreduction when exposed to sunlight in the presence of reducing agents such as alcohol or formic acid. When ammonium carbonate is added to a photochemically reduced solution of uranyl nitrate or formate, a green precipitate is obtained in a fine state, which on washing turns dark green. Analysis of the freshly prepared moist compound was carried out after ammonia had been tested for and found to be absent. The uranium to carbonate ratio was found to be 1 : 1 by semimicro-combustion method. The water content was found to vary from time to time due to the moist compound being used for analysis. The total uranium content was estimated as U_3O_8 and also by the Jones reductor method. That all the uranium exists as uranium (IV) was proved by directly titrating the solution of the compound in 2 *N* sulphuric acid against standard potassium permanganate. On account of the extreme proneness of the compound to oxidation and hydrolysis at high temperature, and also our lack of equipment for isothermal dehydration at low temperature, it has not been possible to get the compound either in the anhydrous state or in a definite state of hydration.

The isolation of uranium (IV) oxycarbonate offers a new route for the syntheses of uranium (IV) compounds[3]. Investigations are proceeding with the view of preparing some new compounds of uranium as well as the dioxide[4].

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[1] Miller, P. D., Pray, H. A., and Munger, H. P., Rep. A.E.C.D. (1949).

[2] Chernyaev, I. L., Golovnya, V. A., and Ellert, G. V., *Zhur. Neorg. Khim.*, 1, 35, 2726 (1956).

[3] Sahoo, B., and Patnaik, D., *Curr. Sci.* (in the press).

[4] Sahoo, B., and Patnaik, D., *Curr. Sci.* (in the press).

Photolysis of 1 : 3-Dichlorotetrafluoroacetone

THE primary act in the photolysis of acetone at room temperature has been shown by a number of workers to be the production of an acetyl and a methyl radical[1]. It is believed that the photolysis of hexafluoroacetone is a simpler process in which only trifluoromethyl radicals and carbon monoxide are formed[2]. Hexafluoroacetone is thus an unambiguous photochemical source of trifluoromethyl radicals, and the close structural similarity of 1 : 3-dichlorotetrafluoroacetone has prompted an investigation into its suitability as a source of difluorochloromethyl radicals.

The near ultra-violet absorption spectrum of 1 : 3-dichlorotetrafluoroacetone shows a broader and more intense band than does that of acetone ($\lambda_{max.}$ 2,900–3,100, $\epsilon_{max.}$ 80, cf. acetone $\lambda_{max.}$ 2,700–2,800, $\epsilon_{max.}$ = 16). A preliminary study of the photolysis of this ketone, at room temperature, using a high-pressure mercury lamp, has indicated that the primary step in this case is also dissociation into two identical radicals and a molecule of carbon monoxide. This conclusion is based upon the absence among the secondary products of reaction of any species which could have arisen from the difluorochloroacetyl radical, for example, tetrafluorodichlorodiacetyl.

The photodecomposition of dichlorotetrafluoroacetone is, however, more complex than that of hexafluoroacetone because the chlorodifluoromethyl radicals formed in the primary step are capable of abstracting chlorine atoms present in the undissociated ketone. Thus in the products of photolysis there appears, in addition to the *sym.*-dichlorotetrafluoroethane formed by the dimerization of chlorodifluoromethyl radicals, a smaller concentration of dichlorodifluoromethane. These two compounds have been isolated from the reaction products by means of gas chromatography, and their identity has been confirmed by comparison of their properties, including their mass spectrometric cracking patterns, with those of authentic samples. A third component of the products, also isolated by gas chromatography, is believed to be 1 : 3-dichlorohexafluoropropane on the basis of its retention volume and mass spectrum, but unfortunately no authentic sample of this material was available to confirm this. Mass spectrometric examination of the reaction mixture has revealed, in addition to mass peaks characteristic of the ketone and of the compounds named above, small mass peaks which can only arise from 1 : 4-dichlorohexafluoromethyl ethyl ketone, and from 1 : 4-dichloro-octafluorobutane. These overall reaction products may be understood on the basis of the following reaction scheme :

$$
\begin{aligned}
&ClCF_2COCF_2Cl \xrightarrow{h\nu} 2\,CF_2Cl— + CO\\
&CF_2Cl— + ClCF_2COCF_2Cl \rightarrow CF_2Cl_2 + ClCF_2COCF_2—\\
&CF_2Cl— + CF_2Cl— \rightarrow ClCF_2CF_2Cl\\
&CF_2Cl— + ClCF_2COCF_2 \rightarrow ClCF_2COCF_2CF_2Cl\\
&ClCF_2COCF_2CF_2Cl \xrightarrow{h\nu} CF_2Cl— + ClCF_2CF_2— + CO\\
&ClCF_2CF_2— + CF_2Cl— \rightarrow ClCF_2CF_2CF_2Cl\\
&ClCF_2CF_2— + ClCF_2CF_2— \rightarrow ClCF_2CF_2CF_2CF_2Cl
\end{aligned}
$$

The excited states involved in the primary decomposition of the ketone may be quenched by the ketone and by other molecules. The behaviour of the ketone in this respect resembles closely that of hexafluoroacetone. A further reaction product that is to be anticipated from the free radicals involved in this system is 1 : 6 dichloro-octafluoro-hexa-2,5-dione, which may be formed by the combination of two chlorotetrafluoro-acetonyl radicals. Although no evidence for the formation of this compound has been obtained, this does not exclude the possibility that it occurs in the products of reaction, since its observation may be made difficult by its anticipated relatively low volatility.

The proportion of dichlorodifluoromethane produced in the photolysis increases with temperature, and further study of this effect will give the energy of activation of the abstraction process.

Where radicals of two species, R_1 and R_2, are involved in a reaction system, these may combine in the following ways :

$$
\begin{array}{llll}
R_1 + R_2 \longrightarrow & \text{products :} & \text{rate constant} & k_{12}\\
R_1 + R_1 \longrightarrow & ,, & ,, \quad ,, & k_{11}\\
R_2 + R_2 \longrightarrow & ,, & ,, \quad ,, & k_{22}
\end{array}
$$

The quantity $\phi^2 = k^2_{12}/k_{11}k_{22}$ is a measure of the preference of unlike radicals to combine. For the chlorodifluoromethyl and 2-chlorotetrafluoroethyl radicals in the present system it has been possible to estimate the rates of accumulation of the ethane, propane and butane, which are formed by these processes, from the mass spectra of reaction mixtures photolysed to small extents. These give $\phi = 2{\cdot}2$, which is about the value required by the simple collision theory of reaction-rates, and is a value close to those tabulated recently for a number of free radicals[3].

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[1] Davis, W., *Chem. Revs.*, **40**, 201 (1947).

[2] Ayscough, P. B., and Steacie, E. W. R., *Proc. Roy. Soc.*, A, **234**, 476 (1956).

[3] Trotman-Dickinson, A. F., Ann. Rep. Chem. Soc., **55**, 39 (1959).

Identification of the Structure of Non-volatile Organic Substances by Gas Chromatography of Pyrolytic Products

AT the Third International Symposium on Microchemistry in Birmingham, Prof. F. Feigl reported on the combination of coloured spot reactions with the pyrolysis of high molecular weight substances for identification purposes[1]. I wish to point out some new possibilities of this type of analysis in the study of the constitution of non-volatile or poorly volatile substances, combining pyrolytic reactions with gas chromatography.

Gas chromatography has been commonly used in the analysis of the products of pyrolytic processes. The work of Desty *et al.*[2] in following the function of internal combustion engines or of Kyryacos *et al.*[3] on the composition of the cool-flame products will be remembered. Recently, gas chromatography has been used for the analysis of substances by the examination of the functional groups split off, the determination of alkoxy-groups[4-6], for example. More recently, a short report on the analysis of pyrolytic products of some polymers[7,8] appeared in the literature.

The principle of the method proposed is very simple. The substance is submitted to controlled pyrolysis in a stream of carrier gas and the products produced are separated and identified by gas chromatography. It can be supposed that the composition of the pyrolytic products will bear a close relation to the structure of the original substance if the pyroly-

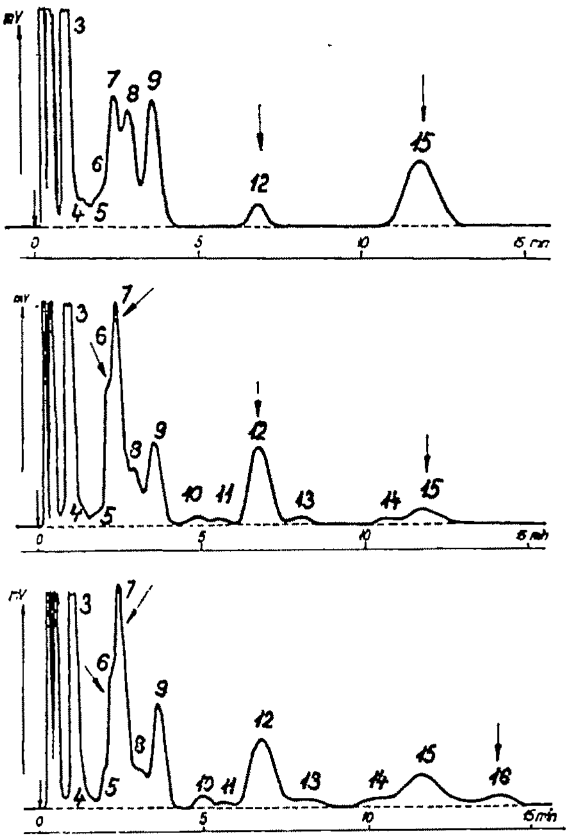

Fig. 1. Chromatograms of the pyrolytic products (C_1–C_6 hydrocarbons) of oils (olive, hardened olive and coconut oil). Fractions 1–4, C_1 + C_2–C_4 hydrocarbons, fractions 5–9, C_5 hydrocarbons, fractions 10–16, C_6 hydrocarbons (squalane, temperature, 40° C.)

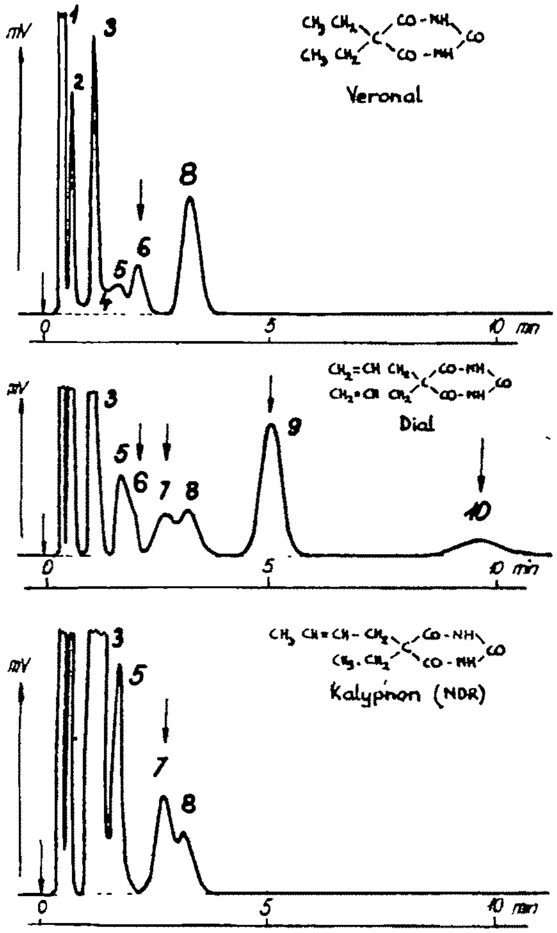

Fig. 2. Chromatograms of pyrolytic products in the area of C_1–C_6 hydrocarbon fragments of sodium salts of barbituric acid derivatives. Fraction 1, C_1 and C_2 hydrocarbons, fraction 2, C_3 hydrocarbons, fractions 3 and 4, C_4 hydrocarbons, fractions 5–8, C_5 hydrocarbons, fractions 9 and 10, C_6 hydrocarbons (squalane, temperature, 40° C.)

sis can be hindered from occurring as a complex reaction of the original substance and the decomposition products being formed. It is necessary in order to achieve a satisfactory solution to have sufficiently extensive measurements of the chromatographic spectra of the various decomposition products and to use an instrumentation technique which allows working with microgram quantities of substances.

Our experiments were made with a high-temperature chromatograph manufactured by the firm (National Plant) Laboratory Instruments, Prague, which is equipped with a flame ionization detector, circulation air thermostat operating up to 350° C., and a recorder with a range of 0·2 mV. Squalane (30 per cent w/w on 'Celite 545') was used as stationary phase, nitrogen as carrier gas, and hydrogen served to maintain the flame. The substance was put on to the column in a quantity up to 0·1 mgm. directly or from an ethereal solution, on a platinum wire of 0·1 mm. in diameter, shaped in spiral form, 15 mm. in length and 1·5 mm. in diameter. By means of an electric time-switch reproducible heating of the platinum wire (up to 800° C.) for a precisely adjusted time (0·1–5·0 sec.) can be attained.

Experiments made with various plant oils and animal fats showed a surprising reproducibility of the chromatographic spectrum quality as well as a quantitative representation of the components, even with relatively large variations of the platinum wire temperatures and the heating time. This situation occurs—and this must be emphasized—only on using minute quantities of substances, 10–50 µgm., when an instantaneous removal of the pyrolytic products from the decomposition area takes place. The pyrolysis of substances with relatively small molecules (for example, fats, steroids) tends to be an 'isothermal' action and the molecule fragments are the result of the power balance within the molecule at this decomposition point and the equilibrium of radicals produced. In the case of pyrolysis of macromolecules such as rubber, etc., the qualitative reproducibility of chromatographic spectrum remains similar, but the quantitative composition is not so consistent.

Fig. 1 shows the pyrolytic spectrum of the C_1–C_6 hydrocarbon products which are formed by the pyrolysis of olive, hydrogenated olive and coconut oils. An obvious qualitative and quantitative shifting of the components in the chromatograms with a change of the chemical nature of the oil is apparent. Other oils, such as linseed oil or castor oil, also exhibit qualitatively and quantitatively specific pyrolytic spectra.

In order to clarify the relation between the composition of the decomposition products and the structure of the original substance, the pyrolysis products of a series of simple structurally related barbituric acid derivatives were studied. In Fig. 2 the chromatograms of three of the fourteen barbiturates studied can be seen. Fragments of the molecules present are always qualitatively as well as quantitatively highly specific. Not only can the group type of a soporific be differentiated, but also its

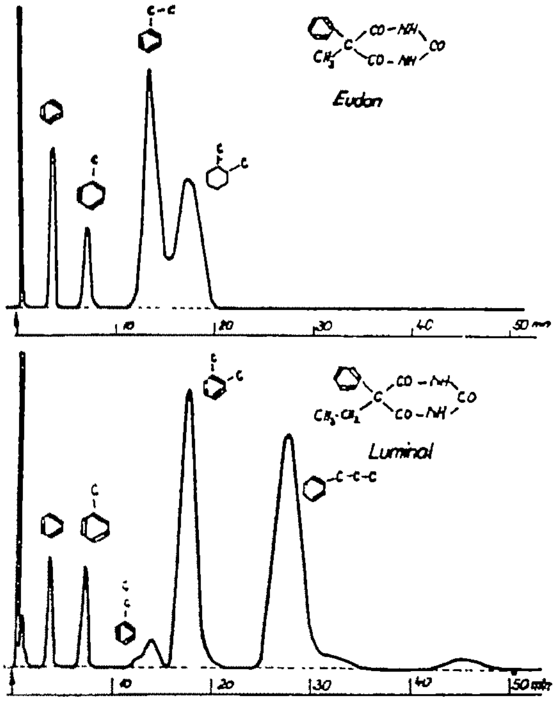

Fig. 3. Chromatogram of the pyrolytic products of the sodium salts of barbituric acid phenyl derivatives in the area of aromatic hydrocarbon fragments (squalane, temperature, 100° C.)

structure can be exactly determined. Experiments made *in vivo* supported the opinion that it would be possible to prove the structure of a barbiturate, after being taken, not only in the stomach content but also in the urine.

The pyrolytic procedure gives not only primary decomposition products. In the decomposition process radicals are formed which react in a characteristic way and form new components indicative of the structure of the original radicals. In Fig. 3 a spectrum of fragments in a range higher than benzene can be seen which shows components that are formed necessarily by the recombination of radicals.

A further improvement permits the use of various chromatographic column fillings for a double or multiple proof of the structure of the pyrolytic substances, the choice of various carrier gases for inducing structural changes in the decomposition products being formed and a temperature change of the chromatographic column (Figs. 2 and 3) for the exposure of a suitable part of the chromatographic spectrum.

This technique, particularly the gas chromatography, has considerably extended analytical potentialities in the field of the poorly volatile and non-volatile substances and is applicable to oils, alkaloids, steroids and other similar natural substances, artificial dyestuffs, drugs and other synthetic materials. Further, it serves as a basis for the development of rapid inquiry methods for medicinal and forensic chemistry, toxicology, biochemistry and biology, and is obviously useful in proving and verifying the structure of chemical individuals. Studies outlined in this communication will be published elsewhere in detail.

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[1] Feigl, F., Proc. Symp. Microchemistry, Birmingham, 1958 (Pergamon Press, London, 1959).
[2] Desty, D. H., Warman, T. J., and Whyman, B. H. F., Symp. Vapour Phase Chromatography, London, 1956, 346 (Butterworth, 1957).
[3] Kyryacos, G., Menapace, H. R., and Boord, C. F., *Anal. Chem.*, 31, 2, 222 (1959).
[4] Vertalier, S., and Martin, F., *Chim. analytique*, 40, 3, 80 (1958).
[5] Kratzl, K., and Grubner, K., *Monatschr.*, 89, 45, 618 (1958).
[6] Springer, H., and Market, T., *Mikrochim. Acta*, 122 (1959).
[7] Lehrle, R. C., and Robb, J. C., *Nature*, 183, 1671 (1959).
[8] Radell, E. A., and Strutz, H. C., *Anal. Chem.*, 31, 1890 (1959).

BIOCHEMISTRY

Effects of the Method of Tissue Preparation on the Assay of Tissue Enzyme Activities

THERE has recently been considerable interest in comparative enzyme-levels of various tissues of different species in health and disease. It is evident that the quantitative determination of enzyme activity in tissues is dependent not only upon the method used for measuring the activity, but also upon the method used for preparation of the tissue for analysis. Various factors, such as the intracellular localization of the enzyme, the availability of coenzymes and activators, and the permeability of both cellular and particulate membranes of different tissues to various substrate and product molecules, play a part in the effect of the preparation of the tissue prior to enzyme assay.

As a general principle, the method of choice for the preparation of tissue should be that by which maximum enzyme activity is obtained, that is, the method of preparation should not cause appreciable loss of enzyme activity, and should remove all membranes limiting the rate of the enzyme reaction. The assay medium should have optimal composition and contain any known coenzymes and activators of the enzyme studied. If careful consideration is not given to these matters significant discrepancies may appear in the work of a single laboratory or in the comparison of results obtained by different investigators.

In the course of some recent studies on the activity of alkaline phosphatase in the kidney of a patient with hypophosphatasia[1], a considerable discrepancy was noted between the results obtained using frozen-dried 16μ sections[2] and those obtained with aqueous extracts of tissue minces[3-5]. In order to investigate the cause of the discrepancy, alkaline phosphatase activity was determined by Lowry's method[6] in rabbit kidney cortex and liver tissue prepared for analysis by various methods. Also determined was lactic dehydrogenase activity by means of a fluorometric method[7].

Five methods of sample preparation were used: frozen-dried sections, crushed frozen-dried tissue, homogenate, frozen-dried homogenate, and minced tissue. The homogenate technique was included because it is the most widely used method of tissue preparation for enzyme assays. The crushed frozen-dried tissue method and the frozen-dried homogenate technique were included because they offer the possibility of long-term storage of the preparations. To obtain frozen-dried sections the tissue was frozen in liquid nitrogen and sections 16μ thick were cut at −20° C. The sections were then frozen-dried, weighed, and placed directly in the substrate medium. For the crushed frozen-dried tissue, the tissue frozen in liquid nitrogen was crushed in a pre-cooled stainless steel mortar by a blow of a hammer on the pestle. Part of the powder was inserted into a previously weighed cold 13 mm. × 100 mm. 'Pyrex' tube, weighed, and lyophilized. The tube was then stoppered and stored at − 35° C. until used. Prior to analysis, the powder was reconstituted with enough water to make it 5 per cent on a wet-weight basis. For the homogenate a 5 per cent aqueous homogenate was made in a Potter–Elvehjem tissue grinder using a Kontes Glass Co. stainless steel pestle (clearance 0·009–0·011 in.) and then a Kel-*F* pestle (clearance 0·004–0·006 in.). The homogenization was continued long enough to pass all the tissue up and down a few times. To obtain frozen-dried homogenate, 1 ml. aliquots of the homogenate were shell frozen in 13 mm. × 100 mm. 'Pyrex' tubes and lyophilized. The tubes were stoppered and stored at − 35° C. until used. The powder was reconstituted with 1 ml. water prior to analysis. In the case of the mince, the tissue was finely minced with scissors and suspended in enough water to make it 5 per cent on a wet-weight basis. The suspension was covered with toluene and stored at 4° C.

The results are summarized in Table 1, upper part. Frozen-dried tissue sections showed both alkaline phosphatase activity and lactic dehydrogenase activity 2–8 times greater than any other preparation. The enzyme activity of frozen-dried homogenate in most instances showed somewhat greater activity than crushed frozen-dried tissue, homogenate or minced tissue. Homogenate and minced tissue stored

Table 1. ALKALINE PHOSPHATASE AND LACTIC DEHYDROGENASE IN LIVER AND KIDNEY CORTEX OF RABBIT

Treatment	Alkaline phosphatase		Lactic dehydrogenase	
	Liver	Kidney cortex	Liver	Kidney cortex
Frozen-dried section	100 ± 3·5 (10)	100 ± 3·6 (8)	100 ± 4·8 (7)	100 ± 5·3 (8)
Frozen-dried homogenate	35 ± 1·4 (3)	33 ± 0·9 (3)	44 ± 1·4 (3)	37 ± 2·0 (3)
Crushed frozen-dried	18 ± 1·6 (2)	—	38 ± 4·8 (2)	—
Homogenate, 1*d*, −35° C.	17 ± 0·6 (2)	29 ± 0·9 (2)	29 ± 0·8 (2)	32 ± 1·4 (2)
,, 7–8*d*, −35° C.	18 ± 0·8 (2)	29 ± 2·3 (2)	27 ± 3·7 (2)	30 ± 3·5 (2)
,, 7–8*d*, +4° C.	28 ± 1·0 (2)	45 ± 1·8 (2)	24 ± 4·9 (2)	26 ± 2·6 (2)
Mince, 1*d*, + 4° C.	20 ± 0·6 (3)	33 ± 1·9 (3)	25 ± 0·4 (3)	32 ± 1·6 (3)
,, 7–8*d*, + 4° C.	35 ± 1·6 (3)	36 ± 1·8 (3)	12 ± 1·7 (3)	25 ± 2·2 (3)
Fresh homogenate :				
in water	100 ± 2·7 (3)	100 ± 3·2 (3)	100 ± 5·0 (3)	100 ± 4·2 (3)
in PO_4—BSA*	83 ± 8·4 (3)	60 ± 3·7 (3)	116 ± 2·9 (3)	90 ± 1·4 (3)
Frozen homogenate, 1*d*, −35° C.				
in water	96 ± 5·6 (3)	113 ± 4·5 (3)	112 ± 2·5 (3)	99 ± 1·8 (3)
in PO_4—BSA*	75 ± 1·3 (3)	62 ± 2·3 (3)	99 ± 3·4 (3)	83 ± 5·8 (3)

* 0·1 *M* phosphate buffer, *p*H 7·4, containing 0·05 per cent bovine serum albumin.

All activities based on dry weight; activities in upper part of table expressed in percentage of the activity of a frozen-dried section of the same tissue, in lower part in percentage of the activity of the fresh homogenate in water. Standard errors of the means shown with number of samples in parentheses.

for 7–8 days showed slightly greater alkaline phosphatase activity and slightly lower lactic dehydrogenase activity than homogenate and minced tissue stored for only one day. In the case of alkaline phosphatase, a very stable enzyme, the rate of solubilization of the enzyme into the extracting fluid is apparently higher than the rate of denaturation, while in the case of lactic dehydrogenase, a less stable enzyme, the rate of denaturation is apparently faster than the rate of solubilization. The high results obtained with frozen-dried sections can probably be explained by the rapid and complete rupture of cell membranes which occurs when the section is put into contact with aqueous substrate. Microscopically this process has been observed as a very rapid swelling and soaking of the dry tissue at the moment of immersion.

In another experiment homogenization was carried out in 0·1 *M* phosphate buffer (*p*H 7·4), containing 0·05 per cent bovine serum albumin, instead of in water. In the same experiment fresh homogenates were compared with those kept at − 35° C. for one day. The results are given in Table 1, lower part. No increase in the activity of either enzyme was noticed by replacing water with the phosphate–albumin medium. In the case of alkaline phosphatase a slight inhibition occurred, probably due to a mass law effect. No significant differences existed between the fresh homogenates and the homogenate which had been frozen and kept at − 35° C. for one day.

It appears that the highest results for both enzymes were found in the frozen-dried sections, while the frozen-dried homogenate method seems to be the next best technique. It is probable that differences in enzyme activity, depending on the method of preparation of tissues for analysis, will also be found for other enzymes. It appears evident that certain discrepancies in comparative tissue enzyme studies can be avoided if cognizance is taken of the critical importance of the methods of preparation of tissues for enzyme assay and the need for careful standardization in each study.

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[1] Rosenthal, I. M., Bonting, S. L., Hogan, W., and Pirani, C. L., *A. Med. Assoc. J. Dis. Child.* (in the press).

[2] Bonting, S. L., Pollak, V. E., Muehrcke, R. C., and Kark, R. M., *Science*, **127**, 1342 (1958).

[3] Bodansky, A., *J. Biol. Chem.*, 101, 93 (1933).

[4] Woodward, H. Q., *Cancer Res.*, **2**, 497 (1942).

[5] Woodward, H. Q., *Cancer*, 9, 352 (1956).

[6] Lowry, O. H., Roberts, N. R., Wu, M. L., Hixon, W. S., and Crawford, E. J., *J. Biol. Chem.*, **207**, 19 (1954).

[7] Lowry, O. H., Roberts, N. R., and Kapphahn, J. T., *J. Biol. Chem.*, **224**, 1047 (1957).

Formation of Carotene *in vitro*

RESULTS presented in this communication indicate that both β-hydroxy β-methylglutarate and mevalonate may be considered precursors of carotene formation in an *in vitro* extract obtained from mycelial mats of the mould *Phycomyces blakesleeanus*. This extract, capable of synthesizing carotene, can also be used for establishing the existence of a back reaction from the mevalonate to β-hydroxy β-methylglutarate, for brevity referred to as glutarate.

The sequence of conversions is, however, undoubtedly similar to those previously proposed for carotenoids[1–4] and sterols[5]. In the initial stages of the formation of isoprenoid compounds, at least, there is an identity in the 5 and 6 carbon intermediates.

Previous work has shown that glutarate and mevalonate are used with considerable efficiency in the synthesis of carotenoids in tomatoes, tomato and carrot homogenates, and by growing cultures of *Phycomyces*[1–4]. Goodwin has reviewed evidence that glutarate is a direct precursor of mevalonate in plant systems[4]. However, the experimental conditions in growing plants were such that enzymatic breakdown to acetate could occur, and thus mevalonate might arise independently.

In a series of tests, a cell-free extract has been obtained from mycelial mats of *Phycomyces* which is capable of synthesizing labelled carotene from acetate 2-^{14}C, glutarate 3-^{14}C and mevalonate 2-^{14}C.

The soluble system was isolated from five-day old cultures of *Phycomyces* grown on a medium containing 2·5 per cent glucose, 0·125 per cent leucine, 0·125 per cent asparagine, with added salts and thiamine. The extract was prepared by disintegrating the mycelial mats (about 100 gm. fresh weight) in a loose-fitting tissue homogenizer, at *p*H 7·8 in 0·2 *M tris*-hydrochloric acid buffer which contained in addition nicotinamide, versene, and glutathione, all 0·002 *M*. The homogenate was centrifuged at 4,000*g* for 10 min. to remove gross particles, after which the supernatant was decanted. This was dialysed against 0·002 *M* *tris*-hydrochloric acid buffer for 12 hr. All manipulations were carried out at 4° C. The dialysed material contained 11 mgm. of protein per ml. In an experiment, 4 ml. aliquots were placed in flasks, containing 10 μM of adenosine triphosphate, 1 μM each of diphosphopyridine nucleotide, triphosphopyridine nucleotide and reduced diphosphopyridine nucleotide and 10 μM magnesium sulphate. Substrates in the amount of 5 μM per 4 ml. were added, and the mixture was then incubated for 18–30 hr. at room temperature.

A protein pellet containing the carotene was obtained by centrifuging the test mixture at 10,000*g*. The β-carotene was extracted from the pellet, chromatographed, and the carotene was crystallized with the aid of carrier, washed with hot absolute alcohol and dried as previously described[6]. The carotene was then dissolved in toluene and bleached under a light source rich in ultra-violet for 8 hr. in capped vials. After bleaching, scintillation phosphors ('PPO' and 'popo') were added by injection through the rubber cap of the vial and the resultant solution counted in a Packard 'Tricarb' scintillation counter.

The interconversion of substrates in the supernatant was established as follows. The solution was passed over a 'Dowex 50' column and afterwards over one of 'Dowex 1', on which the free organic acids were absorbed. They were eluted, re-passed over 'Dowex 50', and thus isolated. Phosphorylated derivatives are apparently lost by this procedure, and only the free acids were obtained. The solution was then concentrated and portions were chromatographed on Whatman No. 1 paper, by use of ethanol–ammonia–water (80 : 5 : 15), solvent 1, and *n*-butanol–acetic acid–water (4 : 1 : 5), solvent 2. The paper was then sprayed with bromphenol blue. The activity of the spots was determined by removing strips of the paper containing the visualized spots and counting in sections in a windowless flow counter.

Table 1 lists the total counts added in the different substrates used, and the resultant activities found in the β-carotene. The activity of the substrates was determined by scintillation counting and the counts were adjusted against an internal counting standard to place results on a comparable basis.

For mevalonate, between 1 and 4 per cent of the activity was incorporated into the carotene; for glutarate, about 0·7 per cent; for acetate 2-^{14}C, only a trace. These figures represent lower limits for recovery of activity since the carotene was crystallized.

Table 2 lists the activity of the organic acids isolated from the supernatants. Acetate gives rise to both mevalonate and glutarate, and these are interconvertible. No significance can be attached to the ratios of glutarate : mevalonate activity as there is no information as to the extent to which glutarate may have been diverted into other paths. In addition to well-defined spots for glutarate and mevalonate with R_F values (inadvertently reversed in our earlier paper[4]) of 0·65, 0·59, respectively, for solvent 1, and 0·80, 0·78 for solvent 2, there was an unknown substance showing considerable activity with R_F values of 0·39 and 0·76 in the two solvents.

Table 1. INCORPORATION OF ACTIVITY INTO β-CAROTENE

Substrate	Total c.p.m. added	Total c.p.m. in carotene
Acetate 2-^{14}C	1,100,000	265
Mevalonate 2-^{14}C	170,000	6,997
Mevalonate 2-^{14}C	370,000	3,724
Glutarate 3-^{14}C	187,000	1,296

Table 2. INTERCONVERSION OF ORGANIC ACIDS

Organic acids substrate added	Total c.p.m. found	
	Mevalonate	Glutarate
Acetate 2-^{14}C	202	1,226
Mevalonate 2-^{14}C	(18,170)	4,140
Glutarate 3-^{14}C	2,096	(2,392)

The net increase in carotene in these experiments is usually of the order of 5–10 μgm. The mildest treatment in the homogenizer with removal of unbroken cells by centrifugation seems the most promising lead to improvement of yield.

The foregoing results indicate that both glutarate and mevalonate may be considered as precursors of carotenoids in the system studied, and the evidence substantiates existence of a back reaction from mevalonate to the (hydroxy- methyl-) glutarate.

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[1] Purcell, A. E., Thompson, G. A., and Bonner, J., *J. Biol. Chem.*. **234**, 1081 (1959).

[2] Shneour, E. A., and Zabin, I., *J. Biol. Chem.*, **234**, 770 (1954).

[3] Goodwin, T. W., "Adv. in Enzymol.", 21, 295 (1959).

[4] Chichester, C. O., Yokoyama, H., Nakayama, T. O. M., Lukton, A., and Mackinney, G., *J. Biol. Chem.*, **234**, 598 (1959).

[5] Lynen, F., in "Biosynthesis of Terpenes and Sterols", ed. by Wolstenholme, G. E. W., and O'Connor, M. (Churchill, Ltd., London, 1959).

[6] Chichester, C. O., Nakayama, T. O. M., and Mackinney, G., *J. Biol. Chem.*, **214**, 515 (1955).

Influence of Sorbic Acid on Acetate Oxidation by *Saccharomyces cerevisiae* var. *ellipsoideus*

SORBIC acid (2,4-hexadienoic acid) is widely used as a food preservative. Its effective action against different micro-organisms[1–5] and its harmlessness as a dietary component[6], as well as the ease of its quantitative determination[7–8], and its chemical stability[9] are among the main reasons for its rapid adoption in the food industry.

Sorbic acid has a selective action against catalase-positive organisms[10], and its use as a component of enrichment media for species of *Clostridium* has been proposed[11]. Although the mechanism of its inhibitory action is not well understood, it has been suggested that it probably interferes with the activity of the dehydrogenases in the cell[12].

In the experiments to be described in this communication, we have used a strain of *Saccharomyces cerevisiae* var. *ellipsoideus* which was isolated from fermenting grape juice. The cells were grown on autolysed yeast–dextrose agar. After incubation for some 15 hr. at 30° C., they were harvested and washed twice with 0·033 *M* monopotassium phosphate solution, and finally resuspended in the same solution to an optical density corresponding to a concentration of 2·0 mgm. dry matter per ml. 2 ml. of this suspension were used for each Warburg

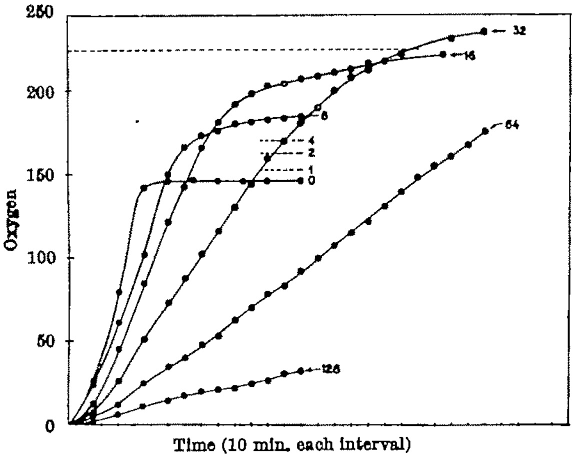

Fig. 1. 4 0 mgm. (dry weight) of cells in each Warburg vessel. Potassium hydroxide in the central cup. 5 μmol. of acetate in all cases. The total amount of sorbate (in μmol.) is indicated at the end of each curve. The upper dotted line represents the theoretical amount of oxygen required for complete oxidation of acetate

vessel, and the oxygen uptake was measured at 30° C. with the conventional technique. Our first experiments showed that there was a small but definite increase of the oxygen uptake over the endogenous by cells in the presence of this compound. This effect was found to be due to an impurity which could be removed by recrystallization from water.

Fig. 1 shows the effect of different concentrations of sorbic acid (added as the sodium salt) on the oxidation of acetate by our strain. The compound has an influence on both the respiration-rate and on the total oxygen uptake by the cells. In the cases where 1, 2 and 4 μmol. of sorbate had been used, only the final values are indicated in the graph. Carbon dioxide determinations by the indirect method showed a respiratory quotient of 1 in all cases.

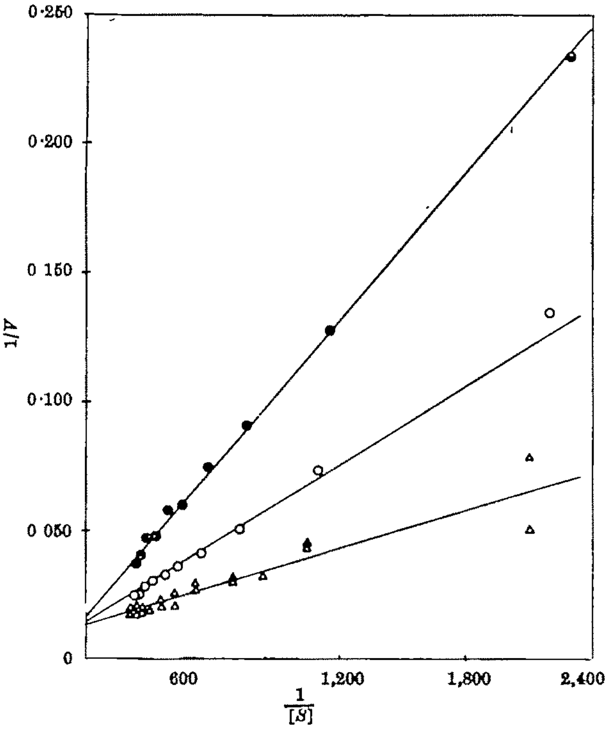

Fig. 2. 4 0 mgm. (dry weight) of cells in each Warburg vessel. Potassium hydroxide in the central cup. The concentration of acetate was varied from 1 to 10 μmol. The straight part of each curve was used for calculating the velocities of respiration. △, No inhibitor; ○, 16 μmol. sodium sorbate; ●, 32 μmol. sodium sorbate

The inhibitory effect of sorbate on the respiration-rate is competitive, as shown by a 'double reciprocal' plot (Fig. 2). Although the exact mechanism of the competition has not been determined, some preliminary experiments seem to indicate that sorbate interferes with the synthesis of citric acid in the cells, probably by way of formation of sorbyl–coenzyme *A*. Sorbic acid also inhibits the acetylation of sulphanilamide by pigeon liver extracts.

The inhibition of acetate assimilation is more difficult to explain. There was no appreciable effect of sorbate on the oxidative phosphorylation by whole cells in the presence of acetate and the inhibitor. Since our manometric experiments were performed under conditions (absence of carbon dioxide in the atmosphere and of combined nitrogen in the medium) which rule out any significant assimilation through the Krebs cycle, it is possible that sorbate could interact with the synthesis of higher fatty acids by the resting cells.

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[1] Phillips, G. F., and Mundt, J. O., *Food Technol.*, **4**, 291 (1950).
[2] Beneke, E. S., and Fabian, F. W., *Food Technol.*, **9**, 486 (1955).
[3] Costilow, R. N., Ferguson, W. E., and Ray, S., *App. Microbiol.*, **3**, 341 (1955).
[4] Costilow, R. N., Coughlin, F. M., Robbins, E. K., and Hsu, Wen-Tah, *App. Microbiol.*, **5**, 373 (1957).
[5] Bell, T. A., Etchells, J. L., and Borg, A. F., *J. Bact.*, **77**, 573 (1959).
[6] Deuel, H. J., Alfin-Slater, R., Weil, C. S., and Smyth, F. H., *Food Res.*, **19**, 1 (1954).
[7] Melnick, D., and Luckmann, F. H., *Food Res.*, **19**, 20 (1954).
[8] Luckmann, F. H., and Melnick, D., *Food Res.*, **20**, 649 (1955).
[9] Alderton, G., and Lewis, J. C., *Food Res.*, **23**, 338 (1958).
[10] Emard, L. O., and Vaughn, R. H., *J. Bact.*, **63**, 487 (1952).
[11] York, G. K., and Vaughn, R. H., *J. Bact.*, **68**, 739 (1954).
[12] Melnick, D., Luckmann, F. H., and Gooding, C. M., *Food Res.*, **19**, 44 (1954).

Low Molecular Weight Organic Base from the Dinoflagellate *Amphidinium carteri*

In the course of an investigation into the nature of the soluble carbohydrate materials released by phytoplankton into their culture medium[1], we found that media from cultures of *Amphidinium carteri*, a dinoflagellate, showed a distinctly blue reaction with N-ethyl carbazole, rather than the usual purple-red colour. The material causing the blue reaction could be removed by dialysis, which also removed the fishy odour characteristic of old cultures of this organism. The dialysate displayed the usual carbohydrate colour reaction.

The substance responsible for the blue reaction, and presumably also for the fishy odour, is very labile. The product originally released into the medium by this organism is somewhat more complex than the dialysable substance responsible for the blue reaction, since samples of filtered medium taken from bacteria-free cultures in the logarithmic stage of growth do not develop the characteristic odour for several hours. The autohydrolysis of the parent compound can be prevented by making the medium slightly acid after filtration, and storing the material at low temperature.

The process of autohydrolysis can be followed by applying the cupric chloride test for aliphatic amines described by Hershenson and Hume[2]. If cold, slightly acid medium is tested, no immediate reaction is visible. After several hours at room temperature a definite positive colour reaction develops. If the parent substance is hydrolysed by refluxing with dilute sodium hydroxide, the test shows an immediate positive reaction.

The dinoflagellate cells contain a substance, probably an enzyme, which can hydrolyse the parent compound very quickly. If cells are damaged in the course of filtration, the fishy odour develops immediately. This hydrolysis can be prevented by the addition to the medium of eserine, an anticholinesterase. The end-product, however, displays none of the chemical reactions of choline. The original product may be an analogue of acetylcholine, since both acetylcholine and acetyl-β-methylcholine behave in the same manner as does the original product in the amine and N-ethyl carbazole tests.

It seems probable that *Amphidinium carteri* releases some analogue of acetylcholine either as a waste product or as a protective device. Such a compound should be physiologically active, and might act as effective protection against zooplankton. The organism itself contains a mechanism for breaking down this compound. This compound or similar compounds may be released by other dinoflagellates. Abbott and Ballantine[3] extracted centrifuged cells of *Gymnodinium veneficum* with acidified methanol and removed a compound the effects of which on mice were similar to those of acetylcholine. An acidified methanol extract of an activated carbon column through which medium from the *Gymnodinium* culture had been passed gave similar results.

This work was carried out at the Osborn Zoological Laboratory, Yale University, New Haven, Connecticut, and the U.S. Fish and Wildlife Laboratory, Milford, Connecticut.

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[1] Guillard, R. R. L., and Wangersky, P. J., *Limnol. and Oceanogr.*, 3, 449 (1958).

[2] Hershenson, H. M., and Hume, D. N., *Anal. Chem.*, 29, 16 (1957).

[3] Abbott, B. C., and Ballantine, D., *J. Mar. Biol. Assoc. U.K.*, 36, 169 (1957).

Enzymic Synthesis of L-Aspartate-4-^{14}C

As the use of radioactive isotopes in research has been extended, there has been an increasing demand for specifically labelled metabolites of high activity. At present the synthesis of such compounds by purely chemical means is often difficult, if not impossible, particularly when only the biologically active isomer is required. While enzymic synthesis is recognized to be desirable in principle, some workers may be deterred by the difficulties involved in preparing the required enzymes to what is considered to be the necessary state of purity.

What may be overlooked is that it is frequently possible to isolate an enzyme biochemically rather than physically by adding to an adequately dialysed mixture only that combination of substrates, coenzymes and co-factors which is necessary for its activation. In these circumstances it may prove possible, as in the present instance, to use crude (and therefore more easily prepared) enzymes to effect specific syntheses.

Previous work[1] has indicated that L-aspartate-4-^{14}C can be synthesized enzymically *in vitro* by combining the reactions catalysed by phosphopyruvate carboxylase[2] and glutamic–aspartic transaminase. The overall reaction is:

$$\begin{aligned}&CH_2:COPO(OH)_2{\cdot}COOH + {}^{14}CO_2 + H_2O + \\ &\qquad HOOC{\cdot}CH_2.CH_2{\cdot}CHNH_2{\cdot}COOH \rightarrow \\ &HOO^{14}C{\cdot}CH_2{\cdot}CHNH_2{\cdot}COOH + \\ &\qquad HOOC{\cdot}CH_2{\cdot}CH_2{\cdot}CO{\cdot}COOH + H_3PO_4 \end{aligned} \quad (1)$$

Recently, experiments have been undertaken in this laboratory in order to determine whether this accurately represents the reaction involved.

The enzymes required for the synthesis were prepared by grinding 100 gm. of *Kalanchoë crenata* leaves in 100 ml. of 0·625 per cent 2-amino-2-hydroxymethylpropane-1 : 3-diol (*tris*). The protein present in the clear supernatant after centrifugation at 20,000*g* was precipitated by making 100 per cent w/v with ammonium sulphate. The precipitate was redissolved in 30 ml. of 1 per cent *tris* buffer at *p*H 7·5 and dialysed against 3 l. of this solution in the cold for 48 hr. and then against 3 l. of mineral-free water for a further 24 hr.

The reaction was started by the addition of excess phosphoenolpyruvate (50 μmoles) to a stoppered vessel of 7 ml. capacity containing sodium bicarbonate (approximately 22 μmoles at 22·8 μc./μmole), enzyme (5·0 ml.), magnesium chloride (7 μmoles), glutamate (75 μmoles), sodium phosphate (7 μmoles) and *tris* buffer (0·10 ml., 20 per cent w/v at *p*H 7·4). After incubation for 1 hr. at room temperature the reaction was stopped by the addition of formic acid. The acidified reaction mixture was boiled to remove formic acid, and then passed through a column of acid alumina which absorbed all the residual radioactivity. Water was passed through the column until activity appeared in the effluent. Ammonia was then passed through the column until the major part of the activity had been recovered.

Two-dimensional chromatography of the ammonium hydroxide eluate followed by radioautography indicated that radioactivity was restricted to two spots. The first of these corresponded in position to that of added aspartate and contained more than 99 per cent of the activity. The second spot, containing less than 1 per cent of the activity, has not yet been identified but may be oxaloacetate.

When the radioactive eluate from the aspartate spot on two-dimensional chromatograms was incubated with indanetrione hydrate in 'Methyl cellosolve' at *p*H 5 the activity was lost, indicating that the label was restricted to the carboxyl groups[3]. Similarly, the activity was lost as labelled carbon dioxide when the eluate was incubated with an enzyme preparation from *Nocardia*. This enzyme has been shown[4] to catalyse, specifically, the quantitative β-decarboxylation of L-aspartate and this result therefore provides convincing evidence that the labelled substance is in fact L-aspartate-4-^{14}C.

A comparison of the radioactivity of the aspartate fraction and that of standard carbonate-^{14}C (measured with a Geiger–Müller counter under similar conditions) indicated that the level of activity in the aspartate was greater than 400 μc. The amount of aspartate synthesized was determined by spectrophotometric measurements of reduced diphosphopyridine nucleotide oxidized in the overall enzymic reaction:

$$\text{aspartate} + \alpha\text{-oxoglutarate} + \text{DPNH}_2 \longrightarrow \text{malate} + \text{DPN} + \text{glutamate}$$

and found to be approximately 20 μmoles. On the basis of this figure the specific activity of the aspartate was evidently close to that of the bicarbonate used in its synthesis.

These results clearly indicate that virtually all the carbon dioxide fixed is located in the β-carboxyl group of L-aspartate and are consistent with the view that equation 1 is an accurate representation of the reaction.

We wish to acknowledge a gift of *Nocardia* enzyme from Dr. Kerridge, M.R.C. Unit of Microbiology, University of Cambridge, and grants from the University of London Central Research Fund and the Department of Scientific and Industrial Research made to one of us (D. A. W.).

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[1] Walker, D. A., and Ranson, S. L., *Plant Physiol.*, 33, 226 (1958).
[2] Bandurski, R. S., and Greiner, C. M., *J. Biol. Chem.*, 204, 781 (1953).
[3] Nelson, C. D., and Krotkov, G., *Canad. J. Bot.*, 34, 425 (1956).
[4] Crawford, L. V., *Biochem. J.*, 68, 221 (1958).

Decrease in Coenzyme I Content of Spleen after Freezing

During an investigation into the effect of the level of coenzyme I on the response of tissues to hydrocortisone, it was necessary to keep the tissues for at least a day before the coenzyme-level could be estimated. As a supply of solid carbon dioxide ('Drikold') was available it was used to freeze the tissues quickly before storing in a deep freeze.

Table 1. Comparison of Diphosphopyridine Nucleotide (DPN$^+$) Content of Freshly Killed and Frozen Rat Tissues

Tissue	γ-DPN$^+$/gm. fresh weight ± *S.E.*				Percentage decrease in DPN$^+$ caused by freezing
	Freshly killed	No. of estimations	Frozen	No. of estimations	
Diaphragm	256 ± 13·25	4	250 ± 7·14	4	—
Kidney	275 ± 8·75	5	277 ± 11·84	4	—
Liver	236 ± 8·43	8	194 ± 6·12	7	18
Blood*	44·2 ± 2·35	5	36·3 ± 2·16	5	18
Spleen	129 ± 5·19	10	16·8 ± 1·41	12	87

* Blood results expressed as γ-DPN$^+$/ml. whole blood.

The method of extracting the oxidized coenzyme I was that of Glock and McLean[1], except that sodium carbonate was used for neutralizing the acid. The diphosphopyridine nucleotide was then estimated by the fluorescence method[2]. Approximately 100 mgm. of tissue was used for each estimation. Rat tissues investigated were diaphragm, kidney, liver and spleen. It was found that freezing as soon as the tissue was removed from the animal had little or no effect on the content of diphosphopyridine nucleotide except in the case of spleen. As spleen contains a high proportion of blood, this tissue was also investigated (Table 1).

Spleens from two other species—rabbit and guinea pig—were similarly treated and the same type of result obtained from frozen and unfrozen tissue (Table 2). Other available freezing agents—liquid oxygen and alcohol/solid carbon dioxide mixture—were used with rat spleen. Values of 17·8 and 34·0 γ-diphosphopyridine nucleotide per gm. of tissue were obtained respectively.

The buffer method for extracting coenzyme I from tissues of Jedeikin and Weinhouse[3] was tried with rat spleen and the fluorescence method used for the estimation. The average value for unfrozen tissue was higher than that obtained from the other extraction technique due to lower fluorescent blanks. Again, frozen tissues gave low results, the percentage of lowering being similar to that previously obtained (Table 2). When the complete method of Jedeikin and Weinhouse was used with large amounts of rat spleen (600 mgm.), diphosphopyridine nucleotide in extracts from frozen tissue was too small to measure accurately (Table 2).

Thawing frozen tissue before adding to the hot extracting fluid did not increase the value of coenzyme estimated above that of unthawed tissue. Length of freezing did not appear to affect the low results. Tissue which had been quickly frozen and estimated immediately gave results similar to tissues that had been stored frozen for some time. Freezing the neutralized extract from unfrozen spleen in solid carbon dioxide before fluorescence estimation did not lower the coenzyme content.

At first an inhibitor of the fluorescence was thought to be present. The blank obtained from frozen tissues was only 37 per cent of that from unfrozen tissues of the same weight, but known amounts of oxidized diphosphopyridine nucleotide (Sigma grade) added to the extracts or to the extracting acid at the same time as the tissues gave similar recoveries from both fresh and frozen tissues.

It was noticed that the colour of the neutralized extract from frozen tissue was a much darker brown than that from unfrozen tissue. Chromatography,

Table 2. Diphosphopyridine Nucleotide (DPN$^+$) Content of Fresh and Frozen Spleen Tissue from Different Species

Animal	Method of extraction*	Method of estimation*	γ-DPN$^+$/gm. fresh weight ± *S.E.*				Percentage decrease in DPN$^+$ caused by freezing
			Fresh	No. of estimations	Frozen	No. of estimations	
Rabbit	1	2	156	2	12·6 ± 1·26	3	92
Guinea pig	1	2	112 ± 3·13	3	13·6 ± 1·18	3	88
Rat	1	2	129 ± 5·19	10	16·8 ± 1·41	12	87
Rat	3	2	159 ± 4·37	6	20·4 ± 1·39	4	87
Rat	3	3	119	2	—	2	—

* Figures relate to references as given in the text.

using pyridine/water as solvent[4], of these extracts after concentration by freeze drying showed larger brown non-fluorescent streaks in samples from frozen tissue than from fresh tissue, but no clearly defined diphosphopyridine nucleotide spot was detected.

The loss of coenzyme I content on freezing would not appear to be due to enzyme activity as the time for any breakdown is too short and the temperature too low. It also does not appear to be a simple inhibition in the fluorescence estimation. Thus apparently during freezing some physico-chemical phenomenon takes place.

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[1] Glock, G. E., and McLean, P., *Biochem. J.*, 61, 381 (1955).

[2] Ciotti, M. M., and Kaplan, N. O., in "Methods in Enzymology", ed. by Colowick, S. P., and Kaplan, N. O., 3, 896 (Academic Press, Inc., New York, 1957).

[3] Jedeikin, L. A., and Weinhouse, S., *J. Biol. Chem.*, 213, 271 (1955).

[4] Burton, R. M., and San Pietro, A., *Arch. Biochem. Biophys.*, 48, 184 (1954).

ANIMAL PHYSIOLOGY

Water Content of Cod (*Gadus callarias* L.) Muscle

In fatty fish such as herring the protein content of the flesh does not show any regular cycle of changes throughout the year[1,2]. Marked variation is shown by the fat, however, which decreases in times of food scarcity, the flesh showing a corresponding increase in water content. Non-fatty fish such as cod do not behave in this way, and in times of scarcity they draw on their body proteins both for metabolic purposes and for building up the gonads[3]. Water then takes the place of the protein, and in the case of cod artificially starved to the point of death it can increase to as much as 88 per cent of the flesh as compared with about 80·5 per cent in normal fish[4].

Although many determinations of the water content of fish muscle have been carried out, the rather small variations encountered in non-fatty fish landed at commercial ports have rarely been commented on. The following results show, however, that considerable biological information can be obtained from them.

The cod analysed were all obtained from the same ground, about 30 miles south-east of Aberdeen. They were filleted within 12 hr. of being caught, and myotomes 8–16, approximately (counting from the anterior end of the fillet) were dissected out free from myocommata (connective tissue) and placed in weighed basins, approximately equal weights of material being taken from each fish in a sample. Water was determined by drying in an oven at 100° C. for seven days.

Three size-groups of fish were used: (1) 20 in. long or less (sexually immature); (2) close to 30 in. long (about the size when they become mature); (3) 36–42 in. long (mature). It was difficult to get fish in the latter group at certain times of the year, and each sample consisted of pooled material from only 1–8 such fish. Material from medium-sized fish was obtained from 10 individuals, while 20 individuals were used for the 'small' group.

The results (Fig. 1) show that the water content is affected by the size of the fish as well as by the season, the maximum in March corresponding with the period of spawning. Not only is tissue hydration greater in larger fish than in smaller, but in addition it lasts longer (about 3 months). The smallest fish are affected for just one month, recovering in April, and it is likely that this effect is due solely to starvation, since the fish are immature. The findings are in accordance with previous observations[3], which showed that the reduction in protein nitrogen was greater in larger fish than in small, owing to the additional need of larger fish to build up gonads on a food supply inadequate even for ordinary metabolism.

There is little doubt that the height of the maximum in Fig. 1, and also its location, depends on the place where the fish were caught, and probably on the year of catching as well. The highest water content that we ever found under natural conditions (87·2 per cent) was in a cod 42 in. long caught off the west coast of Scotland at the beginning of June 1959. The maximum in Fig. 1 is in March 1959, so this isolated observation provides an example of the effect of a different fishing ground.

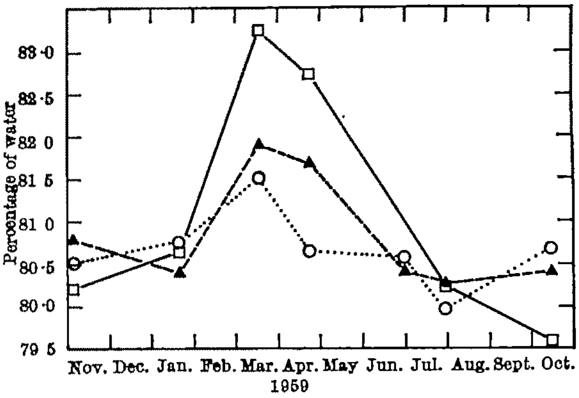

Fig. 1. The water content of cod muscle tissue at different times of the year. ○ · · ○, Small fish; ▲ - - - ▲, medium fish; □ —— □, large fish. Large fish were not obtainable in June

In a recent review of fish composition[5] it was recommended that in order to obtain the complete picture of the variations in a given constituent, analyses should be carried out at monthly intervals for a year on fish of at least two age groups, the fishing ground being the same throughout. From the data in Fig. 1, the recommendation would not appear to have been unreasonable.

The work described in this communication is part of the programme of the Department of Scientific and Industrial Research.

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[1] Lovern, J. A., and Wood, H., *J. Mar. Biol. Assoc. U.K.*, 22, 281 (1937).

[2] Hughes, R. B., *J. Sci. Food Agric.*, 10, 431 (1959).

[3] Ironside, J. I. M., and Love, R. M., *J. Sci. Food Agric.*, 9, 597 (1958).

[4] Love, R. M., *J. Sci. Food Agric.*, 9, 617 (1958).

[5] Love, R. M., "The Physiology of Fishes", ed. by Brown, M. E., 1, 401 (Academic Press, New York, 1957).

Turnover of the Copper and Protein Moieties of Ceruloplasmin

APPROXIMATELY 95 per cent of the plasma copper in both mouse and man is normally present as a moiety of ceruloplasmin[1]. Ceruloplasmin, an α-globulin containing 0·34 per cent copper, exhibits oxidase activity *in vitro* with a variety of substrates[2] and reversibly releases at least half its copper upon reduction[3]. Whether ceruloplasmin plays a part in the transport of copper or acts as an oxidase *in vivo*, however, is not known. In the work reported here, the function of ceruloplasmin in mice was investigated by determining the rate of exchange of its copper moiety.

Human ceruloplasmin isolated from pooled plasma[3] was kindly supplied by Dr. I. H. Scheinberg. The protein moiety of an aliquot of the preparation was labelled with iodine-131 at *p*H 6·8 using the nitrite method described elsewhere[4]. Iodination did not alter the reactivity of the ceruloplasmin with specific rabbit antisera[5] or change its visible spectrum. More than 98 per cent of the radioactivity of the preparation was precipitable in 10 per cent trichloracetic acid. The iodoceruloplasmin was finally screened in Swiss albino mice for 24 hr. to remove any denatured labelled protein, as suggested by McFarlane[6].

Additional aliquots of human ceruloplasmin were labelled with copper-64 using an exchange method[3]. Of the total radioactivity in these preparations, 92–95 per cent was precipitable with rabbit antiserum.

Labelled mouse ceruloplasmin was obtained by injecting mice intraperitoneally with 100 μgm. copper containing 300 μc. copper-64. Serum collected 24 hr. later had a final activity of 1·7 μc. per ml., of which almost 98 per cent was associated with ceruloplasmin electrophoretically. The pooled serum was used as the preparation of mouse ceruloplasmin in these experiments.

Groups of Swiss albino mice 6–7 weeks old were given 0·5 μc. of one of the labelled ceruloplasmins intravenously; this was equivalent to 20 μgm. human ceruloplasmin labelled with iodine-131, 200 μgm. human ceruloplasmin labelled with copper-64, or 40 μgm. mouse ceruloplasmin labelled with copper-64. The mice were assayed in a well-type sodium iodide crystal scintillator. Three or four mice from each group were killed at intervals; serum and various organs were assayed for radioactivity.

The half-life of iodine-labelled human ceruloplasmin, $t_{i\frac{1}{2}}$, as determined by its disappearance from the total body or from serum (Fig. 1), was 0·92 day. The copper-64 in human and mouse ceruloplasmins disappeared from serum with half-lives, $t_{h\frac{1}{2}}$ and $t_{m\frac{1}{2}}$, of 0·75 and 0·85 days, respectively. With human ceruloplasmin at least, copper-64 disappeared more rapidly than the protein moiety was degraded, suggesting a slow exchange of ceruloplasmin copper in the serum; the half-life of this exchange, $t_{\frac{1}{2}}$, was calculated:

$$t_{\frac{1}{2}} = \frac{t_{i\frac{1}{2}} \cdot t_{h\frac{1}{2}}}{t_{i\frac{1}{2}} - t_{h\frac{1}{2}}} = 4{\cdot}1 \text{ days}$$

From 2 hr. following the injection of labelled human ceruloplasmin to the end of the experiment, less than 1 per cent of the serum radioiodide and less than 2 per cent of the serum radiocopper were not precipitable with rabbit antiserum versus human ceruloplasmin[5].

It can be seen from Fig. 1 that the amount of ceruloplasmin in the total body after distribution of the protein in the body fluids was approximately twice the amount present in the serum. Since the average serum ceruloplasmin concentration in these mice was 13 mgm. per 100 ml. serum and the plasma volume was 1·14 ml., the total ceruloplasmin pool was 0·3 mgm. containing 1·0 μgm. copper. With a half-life of 4·1 days, the amount of copper exchanged within the ceruloplasmin pool per day was 0·17 μgm. The mice studied here ingested 45–75 μgm. copper a day in the diet they received; of this, tracer studies with copper-64 indicated that they absorbed a minimum of 4·5–7·5 μgm. copper a day[7]. It is clear that the amount of ceruloplasmin copper exchanged per day was less than 4 per cent of the amount of copper absorbed from the intestinal tract. This would appear to militate against the suggestion that absorption of copper from the intestines is inhibited or controlled by extensive dissociation of ceruloplasmin copper in the intestinal plasma[3]. The small exchange of ceruloplasmin copper would also indicate that ceruloplasmin does not function to transport copper from the intestines unless the intestines synthesize ceruloplasmin. But even the synthesis of ceruloplasmin by one tissue with subsequent release of its copper in other tissues through degradation could result in the transport of only 1·5 μgm. copper per day.

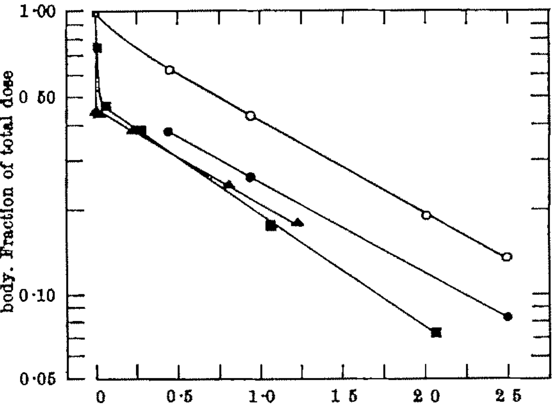

Fig. 1. The disappearance of labelled ceruloplasmins after intravenous injection in mice. From total body: human ceruloplasmin labelled with iodine-131 (○). From plasma: human ceruloplasmin labelled with iodine-131 (●); human ceruloplasmin labelled with copper-64 (■); mouse ceruloplasmin labelled with copper-64 (▲)

On the other hand, the observed exchange in ceruloplasmin copper, although quite small, suggests that ceruloplasmin may act, at least in part, as an oxidase *in vivo*.

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[1] Holmberg, C. G., and Laurell, C.-B., *Acta Chem. Scand.*, **2**, 550 (1948).

[2] Holmberg, C. G., and Laurell, C.-B., *Acta Chem. Scand.*, **5**, 476 (1951).

[3] Scheinberg, I. H., and Morell, A. G., *J. Clin. Invest.*, **36**, 1193 (1957).

[4] Gitlin, D., Janeway, C. A., and Farr, L. E., *J. Clin. Invest.*, **35**, 44 (1956).

[5] Scheinberg, I. H., and Gitlin, D., *Science*, **116**, 484 (1952).

[6] McFarlane, A. S., *Biochem. J.*, **62**, 135 (1956).

[7] Gitlin, D., and Hughes, W. L. (unpublished results).

Effect of Adrenaline and Insulin upon the Oxygen Consumption of Hyperthyroid Rats

It has long been known that the administration of thyroxine to rats, or any other species, is followed by an increase in their basal metabolic rate. In the present experiments rats fed thyroid were found to respond to the injection of adrenaline by a further marked and sustained rise in oxygen consumption. No such increase could be demonstrated in normal control animals. This increased sensitivity of hyperthyroid rats to adrenaline, as reflected in their oxygen consumption, was used to examine the release of adrenaline in insulin hypoglycæmia.

Male and female black-hooded rats (average weight 167 gm.) were used. The addition of thyroxine to their drinking water (0·125 mgm./100 ml.) for seven days before the experiment resulted in a 40 per cent increase in oxygen consumption (from 25 to 35 ml./5 min.). The rats were given no food and only limited water for 18 hr. prior to the experiment. The apparatus used was a modification of the spirometer described by Holtkamp *et al.*[1]; this permitted the oxygen consumption of individual rats to be recorded separately. The rats were allowed to become accustomed to the apparatus for 40–60 min. and thereafter their resting oxygen consumption was determined over a period of 1 hr. The animals were then removed, injected and immediately returned to the spirometer. Following this disturbance there was a transient rise in oxygen consumption; but control experiments in which saline was injected (4 rats, Fig. 1) showed that this was insignificant when compared with the increases following the injection of adrenaline or insulin.

The subcutaneous injection of 50 μgm. of DL-adrenaline into four rats fed thyroid was followed by a considerable rise in the consumption of oxygen (Fig. 1). In four normal rats, the change in oxygen consumption after the injection of the same quantity of adrenaline was insignificant.

The injection of insulin (0·125 unit) subcutaneously into six hyperthyroid rats caused a conspicuous increase in the consumption of oxygen (Fig 2). No such increase could be demonstrated in four normal animals given the same dose of insulin. This rise in oxygen consumption produced by insulin in rats fed thyroid was prevented by the simultaneous intraperitoneal injection of 1·5 ml. of a 30 per cent solution of glucose into a series of eight similar animals (Fig. 2). Any animal that developed hypoglycæmic convulsions was excluded from the series of experiments.

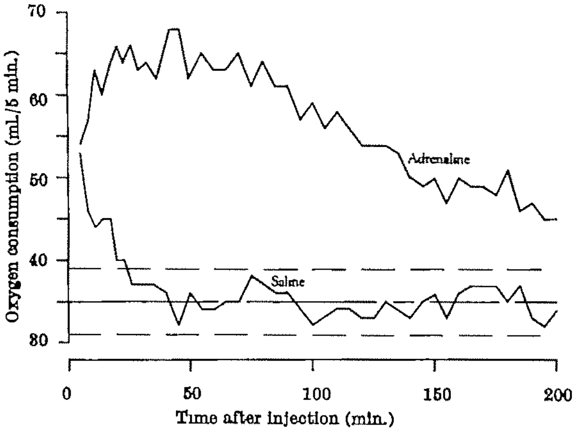

Fig. 1. Effect of adrenaline upon the oxygen consumption of hyperthyroid rats. Upper curve, effect of 50 μgm. of DL-adrenaline in 0 1 ml. saline. Mean of four experiments. Lower curve, effect of the injection of 0·1 ml. saline. Mean of four experiments. The broken lines are drawn two standard deviations from the mean of the resting oxygen consumption

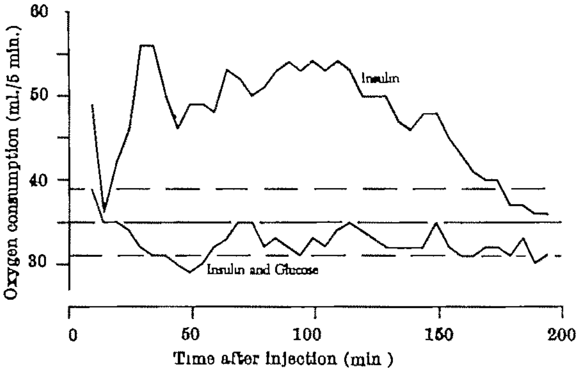

Fig. 2 Effect of insulin upon the oxygen consumption of hyperthyroid rats. Upper curve, effect of 0·125 unit of insulin (subcutaneous injection) and 1 5 ml. saline (intraperitoneal injection). Mean of six experiments. Lower curve, effect of insulin (0·125 unit subcutaneously) and of glucose (1 5 ml., 30 per cent solution intraperitoneally). Mean of eight experiments. The broken lines are drawn two standard deviations from the mean of the resting oxygen consumption

It has been shown[2] that the increased oxygen consumption in the hyperthyroid state could be reduced to normal levels by a total sympathetic block and thereafter restored to the original level by infusion of adrenaline or noradrenaline. As large doses of adrenaline are required to produce even a transient rise in oxygen consumption in normal animals, it was suggested that hyperthyroid animals were unduly sensitive to the calorigenic effects of adrenaline. The sustained and pronounced increase in oxygen consumption following injection of adrenaline into hyperthyroid rats and the insignificant increase produced by the same dose of adrenaline in normal animals, shown in the present experiments, would accord with this suggestion.

The rise in oxygen consumption following the administration of insulin was prevented when the blood sugar was maintained by an injection of glucose; it was therefore inferred that this effect was not a direct consequence of the presence of insulin, but was secondary to the hypoglycæmia produced by this hormone. The demonstration of the release of adrenaline during hypoglycæmia[3] and the evident sensitivity of rats fed thyroid to this agent would imply that the rise in oxygen consumption after the injection of insulin into such animals may be attributed to the secretion of adrenaline. It would appear, therefore, that the oxygen consumption of hyperthyroid rats may be used as an index of changes in the rate of secretion of adrenaline following physiological stimuli.

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[1] Holtkamp, D. E, Ochs, S., Pfeiffer, C. C., and Heming, A. E., *Endocrinol.*, 56, 93 (1955).

[2] Brewster, W. R., Isaacs, J. P., Osgood, P. F., and King, T. L., *Circulation*, 13, 1 (1956).

[3] Cannon, W. B., MacIver, M. A., and Bliss, S. W., *Amer. J. Physiol.*, 59, 46 (1924).

Correlation between the Electrolyte and Water Content of the Organs and Hypertension after Administration of Corticosteroids

IT has been reported that, with renal hypertension, retention of sodium and loss of potassium occurred in the musculature[1–4] and that the same changes in electrolyte metabolism appear after application of hypertension-inducing corticosteroids[5–9]. Therefore these alterations in the inorganic metabolism were considered to be the direct cause of hypertension[10,11]. Tobian *et al.*[12] found an increase in sodium, potassium and water content of the aorta in all forms of experimental hypertension and concluded this to be the cause of hypertension.

In connexion with other researches in the rat, we examined three corticosteroids which influence the electrolyte turnover and the blood pressure in different ways. We also studied their effect on sodium, potassium and water content in the aorta in order to detect which of the changes mentioned above in the electrolyte concentration is related to the blood pressure. The corticosteroids used were deoxycorticosterone acetate (5 mgm./100 gm. body-weight on the first day, then 2·5 mgm.), cortisone acetate (the same dosage) and aldosterone acetate (0·025 mgm./100 gm. body-weight). The treatment lasted 10 days. The daily sodium intake was 75–150 mgm. Sodium, potassium and water content was determined in serum, striated muscle, skin, aorta and in the urinary bladder as another organ containing smooth muscle. With the applied dosage of deoxycorticosterone acetate, the blood pressure remains unchanged in the normal rat (Table 1); as is known, the sodium content of striated muscle is increased and the potassium content is decreased. In the aorta, both sodium and water concentration were increased, whereas the potassium concentration was unchanged.

With adrenalectomized animals the same dosage already produces hypertension. In this case, the sodium as well as the potassium and water content of the aorta is increased; in the urinary bladder an increase in sodium only can be established. It should be emphasized that, by means of the double dosage, hypertension occurs even in normal rats without supplementary supply of sodium chloride.

Cortisone acetate causes hypertension in normal as well as in suprarenalectomized animals. The organs show a depletion of sodium and of potassium, the depletion of potassium being less important than after application of deoxycorticosterone acetate.

It is striking that although the animals survive, the loss of sodium is even more marked than with non-treated adrenalectomized animals. In the aortic wall also the sodium decreases; the potassium, on the contrary, increases strongly.

Aldosterone acetate provokes a retention of sodium in all the organs examined, including the aorta, where the potassium content is almost normal; however, blood-pressure remains unchanged. Thus the experiments show that the variations in sodium concentration of the organs in general or especially in the aortic wall are not conclusive for the production of high pressure.

Table 1

	Blood pressure		Striated muscle			Aorta*			Bladder*		
	Before treatment	After treatment	Na m.Eq.	K m.Eq.	H_2O (per cent)	Na m.Eq.	K m.Eq.	H_2O (per cent)	Na m.Eq.	K m.Eq.	H_2O (per cent)
Normal (10)	128 ± 3·2		23 45 ± 0 2	105·1 ± 0·85	76 0 ± 0·2	265 2 ± 0·8	149 2 ± 0 55	68 8 ± 0 6	310 9 ± 0·85	352 5 ± 0·7	78·4 ± 0·3
Normal + DOCA (7)	131 ± 3 5	135 ± 3 S	30·05 ± 0 75	88·0 ± 0·95	76·0 ± 0·25	299 8 ± 0 55	147 7 ± 0·65	66 5 ± 0 5	304·3 ± 0·9	354 5 ± 0·6	79 3 ± 0 2
Adrenalectomized + DOCA (7)	127 ± 3·2	168 ± 2·5	32 86 ± 0·5	87·4 ± 1·2	76·38 ± 0 3	313 ·8 ± 0·85	171·6 ± 0·75	67·7 ± 0 2	334 0 ± 1·2	358·0 ± 0·8	78·6 ± 0·3
Normal + CA (7)	130 ± 2·8	195 ± 4 2	17·90 ± 0·35	97·8 ± 0·5	75·3 ± 0·5	240·1 ± 0 8	171·5 ± 0·4	68·22 ± 0 2	254·0 ± 0 7	352 1 ± 0 6	78·7 ± 0 2
Adrenalectomized + CA (7)	127 ± 3 2	205 ± 4 0	17·1 ± 0·45	101 7 ± 0·6	75·1 ± 0·45	236 2 ± 0 95	180·5 ± 0·55	62 2 ± 0 25	225 2 ± 0 8	360 5 ± 0·7	77·4 ± 0 2
Normal + AA (5)	132 ± 3·5	130 ± 3 0	25·6 ± 0·4	98·6 ± 0 5	75·29 ± 0·3	294·0 ± 0·9	151 2 ± 0·65	67 8 ± 0 3	345·3 ± 0 9	352·7 ± 1 0	79 2 ± 0·3
Adrenalectomized + AA (5)	128 ± 3·1	122 ± 3·0	30·5 ± 0·4	91·5 ± 0·2	76·1 ± 0·3	318·2 ± 0·85	153·3 ± 0·4	68 5 ± 0 4	351·4 ± 0·95	349·1 ± 0·85	79·7 ± 0·2

* M.equiv./kgm. dry tissue.

DOCA, Deoxycorticosterone acetate; CA, cortisone acetate; AA, aldosterone acetate.

The sodium content in the aorta can be high with normal blood pressure or even low with high blood pressure. On the contrary, the retention of potassium increases parallel with hypertension. It is assumed that the increase of potassium in the aorta wall causes a rise of the tension of the vascular muscle and therefore hypertension. In rats the diameter and length of the large vessels were measured by arteriography. The increase in tension to induce the blood-pressure determined was evaluated.

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[1] Eichelberger, L., *J. Exp. Med.*, 77, 205 (1943).

[2] Laramore, D. C., and Grollman, A., *Amer. J. Physiol.*, 161, 278 (1950).

[3] Leonards, J. R., and Heisler, C. R, *Fed. Proc.*, 11, 247 (1952).

[4] Orbison, J. L., Christian, C. L., and Peters, E., *Arch. Path.*, 54, 185 (1952).

[5] Selye, H., Hall, C. E., and Rowley, E. M., *Canad. ed. Assoc. J.*, 49, 88 (1943).

[6] Thorn, G. W., Engel, L. L., and Eisenberg, H., *J. Exp. Med.*, 68, 161 (1938).

[7] Thorn, G. W., Engel, L. L., and Eisenberg, H., *Bull. Hopkins Hosp.*, 64, 155 (1939).

[8] Gaunt, R., Birnie, J. H., and Eversole, W. J., *Physiol. Rev.*, 29, 281 (1949).

[9] Overman, R. R., *Physiol. Rev.*, 31, 285 (1951).

[10] Braun-Menendez, E., and Covian, M. R., *Rev. Soc. arg. Biol*, 24, 31 (1948).

[11] Braun-Menendez, E., Ciba Foundation Symposium on Hypertension, 238 (Churchill, London, 1954).

[12] Tobian, L., and Binion, J. T., *J. Clin. Invest.*, 33, 1407 (1954). Tobian, L., and Redleaf, P. D., *Amer. J. Physiol.*, 192, 325 (1958); 189, 451 (1957). Tobian, L., and Binion, J. T. *Circulation*, 5, 754 (1952).

Effect of Potassium on Muscle Tension, especially on that of Vascular Muscle

In the previous communication it was shown that in corticosteroid-induced hypertension in the rat, of all the electrolyte shifts, only potassium increase in the aorta wall was characteristic. In striated muscle or in other organs containing smooth muscle, the potassium content either decreases or remains unchanged.

It has been assumed that the increase of concentration of potassium in the vascular muscle causes a tension increase and, as a result, high pressure. Because of this the following experiments were done *in vitro*: (*a*) the effect of potassium on muscle tension; (*b*) qualitative differences concerning the effect of potassium on different muscle types were investigated.

The experiments were especially carried out on carotis strips of cattle, on the musculus rect. abd. of the frog and the rat as well as on the muscle of large intestine of the rat in Tyrode bath, at adequate temperature and with known, variable initial tension. The changes in tension were registered with a transducer fitted perpendicularly to the surface of the examined tissue.

If the potassium content of the Tyrode solution is increased, the tension of the vessel strip increases too. Contrary to Raab's findings[1] the increase of tension can last up to 90 min. according to potassium content and will last as long as potassium is not changed (Fig. 1).

The latent period, the steepness of the tension increase and the total tension depend on the potassium of the bath solution. The maximal tension is attained with a concentration of 60–70 m*M* (Fig. 2). An equilibrium between the bath solution and the extracellular space is established after about 5 min. The increase in tension initiated by potassium and the decrease after removal of it are not influenced by the oxygen content of the bath solution, as is the case with tension caused by electric stimulation or epinephrine. The influence of potassium on striated muscle or on smooth muscle is different. With skeletal muscle as well as with smooth muscle of large intestine only a temporary increase in tension could be produced with the concentrations used. In the muscle of large intestine 30 m*M* potassium first produces an increase in tonus, then a rhythmic fluctuation in tonus.

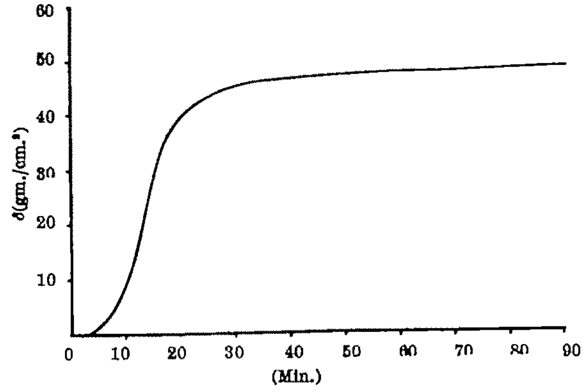

Fig. 1

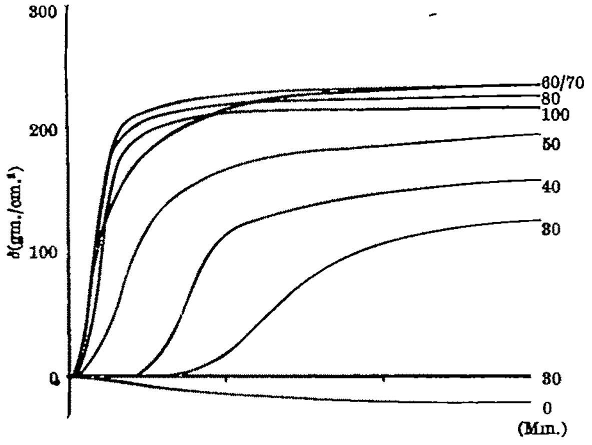

Fig. 2. Course of tension with different concentrations of potassium. Figures on the right indicate m*M* [K]

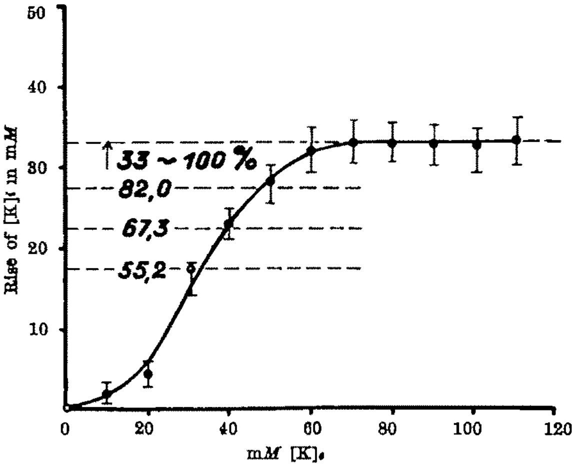

Fig. 3. Abscissa, m*M* in Tyrode solution. Ordinate, m*M* $[K]_i$ increase, after 30 min.

Since on one hand with high-pressure animals the intracellular potassium in the aortic wall is increased, and on the other, as shown, potassium has different effects according to muscle type, the vascular muscle and the muscle of large intestine were examined in regard to their permeability to potassium. The $[K]_i$ was calculated from the total content of K − $[K]_e$. The value of the extracellular space was determined with thiosulphate. As shown in Fig. 3, potassium already penetrates into the vascular muscle when the [K] of the bath solution amounts to 10 m*M* and saturation occurs with a [K] of 60–70 m*M*. If we set the maximum value of the tension increase (Fig. 2) and the $[K]_i$ (Fig. 3) equal to 100, it will be found that the relative tension is proportional to the relative $[K]_i$ (Fig. 3). The course of penetration of potassium into the muscle fibre and of that of tension are similar within a given concentration of the bath solution. No potassium enters the muscle of large intestine of the rat under the same experimental conditions.

It is assumed that two different functional systems are present in the vascular muscle, the contractile system for the transient regulatory tension and the tonic system for the continuous tension. The tension of the contractile system seems to depend on the concentration of potassium in the muscle.

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[1] Raab, E., *Pflügers Arch.*, 215, 651 (1927).

HÆMATOLOGY

Sedimentation of Blood without Anticoagulants

Sedimentation of blood usually occurs only after anticoagulants have been added to prevent clotting and allow time for settling. In devising a technique to study thrombosis *in vitro*[1], an apparatus was made for collecting blood free of tissue thromboplastin and without the addition of anticoagulants. It was incidentally observed that coagulation of blood so collected was delayed sufficiently long for sedimentation to occur. A similar apparatus for collecting blood to study coagulation has been reported by Spaet, Cintron and Kropatkin[2].

Venous blood is drawn directly into a polyvinyl plastic tube through an attached 19-gauge thin-wall, silicone-coated needle. On the end of the tube (opposite the needle) a silicone-coated, three-way stopcock is attached and an additional length of plastic tubing is attached to the stopcock (Fig. 1). As blood flows through the tube, air is pushed out by the advancing column of blood. 2 ml. of blood are allowed to flow out of the tube and are discarded; the end of the tube is clamped and the needle withdrawn from the vein.

No mixing of blood takes place during collection and none of the blood in the tube is exposed to air. By attaching an empty syringe to the stopcock, blood contained in the long section of tubing may be pumped into other containers for further study (Fig. 1). The short section of tubing is clamped off at each end, removed from the stopcock, and hung in a vertical position. The blood in this clamped-off tube will not clot for 1½–2 hr. in most cases and even longer in some instances. Since the blood coagulates so slowly it will settle out rapidly into two distinct layers of plasma and cellular elements (Fig. 2).

The tubing used is a clear polyvinyl chloride plastic tube that measures 0·133 in. in nominal internal diameter (the tubing was kindly supplied by Mead Johnson and Co., Evansville, Ind.). The tubing varies slightly in internal diameter from section to section by volume measurement. 1 ml. fills 10·5–11·4 cm. of tubing. The long section of tubing used is 64 cm. in length and the short section, 28 cm. A total of 10–11 ml. of blood (including discarded blood) usually is withdrawn. The short section of tubing is marked off at 6-cm. intervals, and clamps are attached to separate the blood column into four equal compartments. Sedimentation occurs in all four compartments and the column of clear plasma in the upper compartment is measured at 30 min. Although there usually is some increase in fall after 30 min., most of the sedimentation has occurred by this time and readable values are obtained. The lower three compartments are opened at 1-hr., 1½-hr. and 2-hr. intervals, respectively, to check for the presence of clotting.

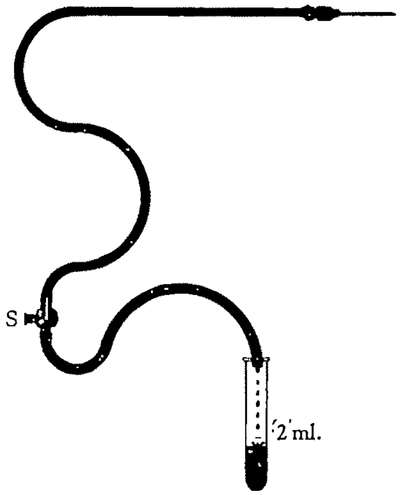

Fig. 1. Diagram of collecting apparatus. (*S*), Three-way stopcock. No suction is required for withdrawing blood. Flow can be controlled by pinching the end of the tube

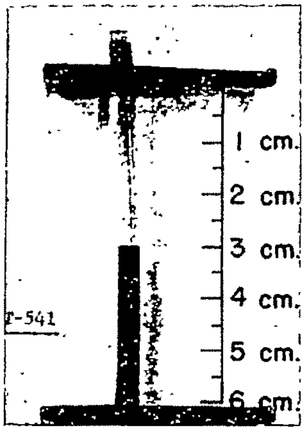

Fig. 2. Sedimentation tube 30 min. after collection of blood from a 52-year-old Negro male with arrested pulmonary tuberculosis. Sedimentation of 3·0 cm. or 50 per cent of the column has occurred. The blood in the upper section beyond the clamp clotted at 45 min. The blood in the enclosed section was not clotted at 2 hr.

Sedimentation-rates have been determined on the blood of 40 male and 71 female patients of the Out-Patient Clinic of the Eugene Talmadge Memorial Hospital of the Medical College of Georgia. Most of the patients were pre-operative and post-operative and a few were prepartum and postpartum cases. The amount of sedimentation in 30 min. averaged 1·8 cm. or 30 per cent of the 6-cm. column in male patients and 1·9 cm. or 31·6 per cent of the column in female patients. In all cases the blood did not clot for 1 hr. or longer. In one instance no sedimentation occurred in 30 min. even though the blood did not clot for 2 hr. The range of sedimentation in 30 min. varied from 0·5 cm. to 4·0 cm. All experiments were done at room temperature, 21–27° C.

Why is coagulation delayed long enough for sedimentation to occur using this method? Several factors are probably important: (1) Silicone-treated metal and plastic tubing are used. Both silicone and plastic provide non-wettable surfaces that retard clotting. (2) Contamination of the blood with tissue thromboplastin is avoided by allowing the first 2 ml. of blood to pass out of the tube during collection. (3) The blood does not make contact with air. According to an early observation of Babington[3], blood collected under olive oil would "enable its red particles to settle more quickly than would otherwise be the case". Müller[4] interpreted this observation to mean that the delayed coagulation was due to exclusion of air. In the method here reported, all air is excluded from the sedimentation tube because air is pushed out of the tube during collection of the blood. The blood in the upper end of the tube beyond the clamp is exposed to air. This blood usually coagulates in 15–45 min. and always coagulates sooner than the blood in the enclosed part of the tube (Fig. 2).

Settling of relatively stagnant blood in blood vessels may be an important factor in the pathogenesis of venous thrombosis, especially in dependent veins of immobilized patients. Fahraeus[5] demonstrated rapid sedimentation of blood in a vein by applying two separated tourniquets on the forearm that was held in a vertical position. In pregnant women or diseased individuals, clear plasma could be obtained in 15 min. by venipuncture just below the uppermost tourniquet. Convincing evidence that settling of blood occurs in horizontal blood vessels in conditions such as stasis has been reported by Harding and Knisely[6]. It is interesting that sedimentation of blood in the apparatus described here was first observed when the tube was horizontal. Settling to the lower side of the tube occurred in a few minutes. Sedimentation of blood as reported here provides a method for studying the suspension stability of stagnant blood *in vitro* without anticoagulants.

The suspension stability of blood is known to be affected by the type and concentration of anticoagulant used in the various sedimentation methods that require anticoagulants[7]. The sedimentation-rate of incoagulable hæmophilic blood can be altered by the addition of heparin or sodium citrate[8]. It is important to know whether or not sedimentation methods using anticoagulants give values paralleling those with the method here described. This method is easily applicable to such a study of both normal and abnormal blood. We are at present using this method to record sedimentation on all specimens of blood used in our *in vitro* thrombosis studies[1], and these results will be reported.

This research was supported in part by a grant from the U.S. Public Health Service, *H*–3973. I wish to thank Miss Marion Hutson and Miss Dorothea Fortson for assisting me in this investigation.

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[1] Chandler, A. B., *Lab. Invest.*, 7, 110 (1958).

[2] Spaet, T. H., Cintron, J., and Kropatkin, M., *J. Lab. and Clin. Med.*, 54, 467 (1959).

[3] Babington, B. G., *Med. Chir. Trans.*, 16, 293 (1830).

[4] Muller, J., "Elements of Physiology", 148 (Lea and Blanchard, Philadelphia, 1843).

[5] Fahraeus, R., *Physiol. Rev.*, 9, 241 (1929).

[6] Harding, F., and Knisely, M. H., *Angiology*, 9, 317 (1958).

[7] Denson, K. W., *J. Clin. Path.*, 12, 262 (1959).

[8] Ham, T. H., and Curtis, F. C., *Medicine*, 17, 447 (1938).

A Fast-moving Hæmoglobin in Hydrops Fœtalis

DURING a study of the hæmoglobins of premature and still-born infants, a fast-moving hæmoglobin was found in a premature baby which clinically showed the symptoms of hydrops fœtalis associated with anæmia, hepato- and spleno-megaly, and which died shortly after birth.

Hæmoglobin analysis of the blood of the fœtus by the paper electrophoretic method as described previously[1] at *p*H 8·6 showed two hæmoglobin spots. One had the mobility of fœtal hæmoglobin, while the other was a fast-moving hæmoglobin, which was much faster than hæmoglobin *A*, slower than hæmoglobin *H* and resembled hæmoglobin *J* and hæmoglobin Fessas and Papaspyrou. At *p*H 6·5, however, *H* and *J* did not separate from hæmoglobin *A*, while the above-mentioned hæmoglobin was clearly separated, and resembled the hæmoglobin of Fessas and Papaspyrou. It was clearly more cathodic than hæmoglobin *H*. It is a pity that we did not have available for comparison the Barth's hæmoglobin, which was described by Ager and Lehmann[2] as being slightly faster than hæmoglobin Fessas and Papaspyrou and faster than hæmoglobin *J* at *p*H 8·6.

The unusual component in the fœtus mentioned above formed the major component, contrary to what has been found up to now in all the cases of newly born infants carrying a fast-moving hæmoglobin component. The amount of alkali-resistant hæmoglobin examined by the method of Singer *et al.* (1951) was found to be 44·3 per cent of the total amount of hæmoglobin.

The symptoms found in the fœtus closely resembled those seen in hydrops fœtalis caused by antagonism between infants and mother's blood.

Also the pathological signs in the grossly enlarged placenta were those seen in hydrops and erythroblastosis fœtalis. There were no indications of antagonism between mother's and infant's blood. However, both parents showed hæmatological abnormalities resembling those found in thalassæmia minor. No pathological hæmoglobin was detected in either parents. The alkali-resistant hæmoglobin was within normal limits.

This is, so far as we know, the first report of a case of hydrops fœtalis associated with the presence of an unusual hæmoglobin.

We wish to thank Prof. Sutomo Tjokronegoro of the Department of Pathology for his co-operation, and Dr. Lie Hong Gie and the nurses of the Obstetric Department of Jang Seng Ie Hospital for their kind help.

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[1] Lie-Injo Luan Eng and Oey Hoey Giok, *Lancet*, i, 20 (1957).

[2] Ager, J. A. M., and Lehmann, H., *Brit. Med. J.*, i, 929 (1958).

PLANT PHYSIOLOGY

Relation of Sugar Content to Frost-Hardiness in Plants

IT has long been said that in various plant cells there is an apparent correlation between the sugar content and frost-hardiness. Because of the difficulties in artificially introducing the appreciable amount of sugar into the plant cell, direct evidence has not yet been obtained to show whether the increase in sugar content is the main cause of the frost-hardiness in plant cells or not.

In artificial frost-hardening of the parenchyma cells in cortex of mulberry tree with a constant length of hardening period, the lower the temperature to which material is exposed, the greater the effectiveness of the hardening treatment becomes. At a definite temperature a relatively longer period of the treatment is more effective than shorter ones within certain limits. In all these experiments, the increase in frost-hardiness of parenchyma cells is closely proportional to that in their sucrose concentration, but the former is not accompanied by any increase of their soluble protein provided that they are artificially chilled[1].

It becomes necessary, however, to distinguish the effect of low temperature itself and that of the increase in sucrose concentration in considering the frost-hardening process in plant cells as a result of artificial chilling. When pieces of twig cut from a mulberry tree in April were gradually desiccated at room temperature (about 20° C.) for 2–3 days, considerable changes[2] took place in the cortical tissues; namely, decrease both in water content (12 per cent per dry weight) and in starch content (32 per cent per dry weight), increase both in sucrose (50 per cent per dry weight or 83 per cent per wet weight) and in the osmotic concentration (0·35→0·53 *M* sodium chloride). When these pieces of twig were frozen at − 10° C. and − 20° C. for 24 hr., the percentage of survival of the parenchyma cells in the twig pieces was 100 and 70, respectively, but that in normal twig (undesiccated) was 25 and 0, respectively. The cells desiccated at room temperature showed greater increase in frost-hardiness as well as in sucrose concentration than the parenchyma cells in the twig pieces which were chilled at 0° C. for 10 days at that time.

Pieces of twig upon which artificial frost-hardening had been effective[1] were kept about 20 days at 15° C. without desiccation (enveloping them with vinyl sheet) in order to use up the starch granules in the parenchyma cells. Afterwards, these pieces of twig were chilled for 10 days at 0° C., but the parenchyma cells in these twigs did not increase in frost-hardiness or in sucrose concentration. Thus, even at a relatively higher temperature (20–25° C.), the increase in sucrose concentration in the parenchyma cells also resulted in rise of their frost-resistance. On the other hand, whenever any chilling in parenchyma cells resulted in no increase in their sucrose concentration, they could not be hardened further by chilling. These results seem to indicate that, at least in the mulberry tree, low temperature itself has no direct effect on the increase in frost resistance; but the increase of sucrose concentration may be a primary factor in frost-hardiness.

Furthermore, some experiments were made to find out whether the increase of sucrose concentration in plant cells directly results in an increase of their frost-hardiness or not. In order to increase the sucrose content in a cell, twigs of gardenia (*Gardenia jasminoides* Ellis) with 2–3 leaves in September and October (the leaves can withstand freezing at − 5° C. for about 16 hr.) were put in the glucose solution (0 7 *M*) at 25° C. for 24 hr. At the same time, the twig of the control was put in water under the same conditions as mentioned above. The osmotic value of the spongy or the palisade mesophyll cells in a leaf of gardenia was 0·7–0·75 *M* (indicated on the basis of sodium chloride). Osmotic concentration, sugar content and degree of frost-resistance in the palisade and spongy parenchyma cells in leaves of the twig treated in glucose solution or in water are shown in Table 1.

Table 1. EFFECT OF GLUCOSE UPTAKE ON FROST-HARDINESS IN LEAF CELLS

Treatment	Osmotic concentration† in cells after treatment	Changes in osmotic concentration in cells after treatment	Sugar content in leaves* (per wet weight (gm.))	Percentage of survival after freezing for 24 hr.	
				at −10° C.	at −15° C.
Water (control)	0 7 *M*	−0·05 *M* (decrease)	0·018	0	0
Glucose solution (0 7 *M*)	1 20 *M*	+0·50 *M* (increase)	0·036	100	80

* Exclusive of nerve fraction.
† Mol. indicated on the basis of sodium chloride.

After the treatment the actual increase of osmotic concentration in the cells amounted to about 20–25 atm., with concomitant considerable rise in their frost-hardiness. Furthermore, some other sugars besides glucose, namely, xylose, sucrose, maltose and raffinose, were also found to be able to penetrate into the cells. In such a case, these sugars showed some protective action against freezing injury in gardenia leaf cells.

It is obvious from these results that the increase of sugar content in a cell results in that of its frost-hardiness.

In addition, polyols (ethylene glycol, glycerol and mannitol) and acetoamide also showed a protective action against freezing in gardenia leaf cells, but inorganic salts did not.

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[1] Sakai, A., *Low Temp. Res.*, Ser. *B*, **15**, 17 (1957).
[2] Sakai, A., *Low Temp. Res.*, Ser. *B*, **17**, 29 (1959).

Damaging Effect of Drying on *Vicia faba* Seeds

IN studies of radiation damage in seeds it has been shown that very dry seeds are more radiosensitive than seeds of medium water content[1,2]. This finding initiated a number of detailed investigations into the radiobiology of very dry seeds to obtain further information on the damaging mechanism of the radiation[3-5]. It has generally been assumed in these studies that drying itself has no harmful effect on the seeds and, for the frequently used barley, this certainly seems to be true according to Caldecott[1], Ehrenberg[2] and Wellington (personal communication). In other species, however, the situation may be rather more complicated. Thus in an earlier paper[6] it has been shown that drying of *Vicia faba* seeds over calcium chloride at 20° C. for 14 days reduces germinability to about 60 per cent and slows down root and shoot growth considerably. Further study of these effects is clearly most important for radiation work in seeds. The effects in *Vicia faba* seeds have, therefore, been investigated in rather more detail.

Seeds of an inbred line of *Vicia faba* var. *minor*, 'Throws M.S. Winter Beans' from Hasler and Co., Ltd., Dunmow, Essex, were used for the experiments. The beans were kept in desiccators in a single layer for periods up to 4 weeks at 20 ± 0·5° C. over the following substances: $NaHSO_4.H_2O$ (laboratory reagent, relative air humidity 52·0 per cent), $CaCl_2.6H_2O$ (analytical reagent, relative air humidity 32·3 per cent), $CaCl_2$ (laboratory reagent, relative air humidity 2·5 per cent).

Water content is sometimes deduced from the conditions and time of storage in desiccators[4]. It was, however, considered necessary to measure the water content of each sample because it is dependent not only upon the time of storage and the drying agent, but also upon the number of seeds and the volume of the desiccator and other factors. The water content was measured after various periods of storage by crushing samples of 10 beans and drying at 105° C. to constant weight. The measured water content of the various samples used in the experiment varied between approximately 12·0 per cent for beans stored in the room to 7·5 per cent after storage for 4 weeks over calcium chloride, the equilibrium water content over this agent being about 4·5 per cent[7].

For germination experiments samples of 30 beans were soaked at $20 \pm 0{\cdot}5°$ C., after pricking the testas, for an initial 16 hr. in aerated demineralized water, and for a further 3 days in aerated tap-water with constant supply (about 12 l./hr.). The number of beans germinated was then counted, the criterion for germination being taken as visible splitting of the testa over the tip of the radicle.

For growth measurements, 20 germinated beans (where possible) from each soaked sample were suspended by means of a perforated 'Perspex' shelf in aerated circulating tap-water (constant supply of about 12 l./hr.) in a controlled-temperature water-bath run at $20 \pm 0{\cdot}5°$ C. After growth for 8 days, root and shoot length were measured. The beans were stored in darkness except for the final pricking, and soaked and grown in darkness except for 5 min. daily at less than 20 ft. candles. These short exposures to light should not affect the ultimate length of the shoot[8].

The scatter diagram for germination percentage in relation to water content (Fig. 1) shows that germination is progressively reduced as the seeds are dried. The correlation coefficient calculated from the results is $+ 0{\cdot}789$, which for normally distributed data would be very highly significant ($P < 0{\cdot}001$). Fig. 2 shows the effect of drying on root and shoot growth. A sigmoid curve would be expected and free-hand curves have been drawn in to assist the eye.

Schoorel[9] has pointed out that it cannot be taken for granted that seeds in general maintain their quality better at low moisture-levels, but that some species from temperate zones as well as a number of tropical species have a relatively high water optimum water-content for storage. Evidently *Vicia faba* belongs to the latter category ; the seeds cannot b dried much below their normal water content withou injury. It may well be that other seeds used in radic biological experiments have similar properties.

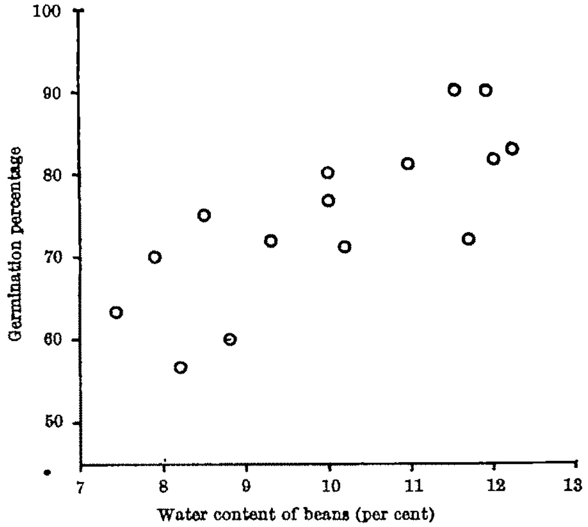

Fig. 1. Germination percentage of *Vicia faba* seeds after soaking for 16 hr. + 3 days, related to water content before soaking

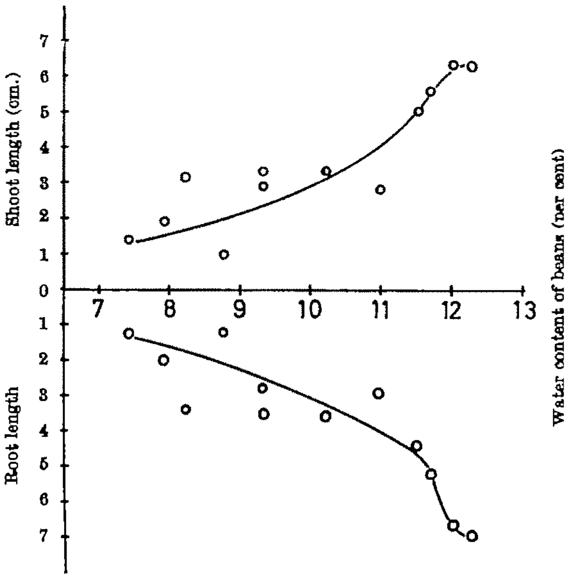

Fig. 2. Root and shoot length of *Vicia faba* after soaking seeds for 16 hr. + 3 days and growing for further 8 days, related to water content before soaking

There appears at present to be no explanation c this harmful effect of drying on seeds. It had nc escaped us that there might be some direct connexio between this phenomenon and the induction b drying alone of free radicles in the testa[7,10]. Thi however, appears to be unlikely, as we have found tha removal of the testa before drying does not appre ciably reduce the final damage.

The results clearly show that in radiobiologic work on dry seeds the effect of drying alone on th seeds may be an important factor to be taken int account in the design and interpretation of exper ments.

We wish to thank Dr. P. Wellington of the Nation Institute of Agricultural Botany, Cambridge, fc discussions of the problems involved, and Pro W. T. Williams of this Department for advice. On of us (W. K.) is also indebted to the Department c Scientific and Industrial Research and the Deutsch Akademische Austauschdienst for a research student ship in Great Britain.

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Dec. 2.

[1] Caldecott, R. S., *Rad. Res.*, 2, 339 (1955); 3, 316 (1955).

[2] Ehrenberg, L., *Botaniska Notiser*, 108, 184 (1955); *Hereditas*, 4 123 (1955).

[3] Caldecott, R. S., Johnson, E. B., North, D. T., and Konzak, C. F *Proc. U.S. Nat. Acad. Sci.*, 43, 975 (1957).

[4] Curtis, H. J., Delihas, N., Caldecott, R. S., and Konzak, C. F *Rad. Res.*, 8, 526 (1958).

[5] Ehrenberg, A., and Ehrenberg, L., *Arkiv för Fysik*, 14, 133 (1958

[6] Klingmuller, W., *Naturforsch.*, 14, b, 268 (1959).

[7] Klingmuller, W., Lane, G. R., Saxena, M. C., and Ingram, D. J. E *Nature*, 184, 464 (1959).

[8] Lane, G. R., and Butler, R. D., *Nature*, 181, 1080 (1958).

[9] Schoorel, A. F., *Het Landbouwkundig Tijdschrift*, 70, 117 (1958 and preprint of Twelfth Internat. Seed Testing Convention, Osl (July 1959).

[10] Klingmuller, W., and Saxena, M. C., *Naturwiss.*, 46, 475 (1959).

HISTOLOGY

Reciprocal Relationship of Phosphorylase and Oxidative Enzymes in Skeletal Muscle

WHILE investigating various enzyme systems in a series of dystrophic and normal human muscles we observed that individual fibres showed a reciprocal relationship between phosphorylase and oxidative enzyme content. Although other workers have investigated the oxidative enzyme systems of normal[1-3] and dystrophic[4] muscle, no observations have been made on the relationship between the two enzymes.

Biopsies were obtained from the lateral head of the gastrocnemius in fourteen cases of progressive muscular dystrophy of the Duchenne type ('pseudohypertrophic' muscular dystrophy) and six normal children. All specimens were immediately quenched in liquid air, stored at − 70° C., and cut within a few hours on a cold microtome at − 20° C. and at 10–15μ.

In an attempt to explain the findings in human muscle a number of different muscles from the rat, pigeon, toad (*Xenopus*) and goldfish were also examined.

The methods employed were those for diphosphopyridine nucleotide and triphosphopyridine nucleotide diaphorases[5]; diphosphopyridine nucleotide-linked β-hydroxybutyrate, *iso*citrate, malate, glutamate, α-glycerophosphate, alcohol and lactate dehydrogenases and triphosphopyridine nucleotide-linked *iso*citrate dehydrogenase[6]; succinate dehydrogenase[7]; cytochrome oxidase[8] and glucosan phosphorylase[9].

With the exception of β-hydroxybutyrate dehydrogenase, all the oxidative enzymes and phosphorylase were active in both normal and dystrophic human muscle. Diphosphopyridine nucleotide-diaphorase and lactate dehydrogenase were the strongest.

In dystrophic muscle the majority of abnormally large fibres (100–200μ) reacted weakly for all the oxidative enzymes and strongly for phosphorylase, while the small atrophic fibres (20–30μ) tended to react strongly for oxidative enzymes and weakly for phosphorylase (Fig. 1*A* and *B*). The normal muscle showed a similar variation (Figs. 2 and 3). Longitudinal sections established the uniformity of reaction throughout the length of a fibre.

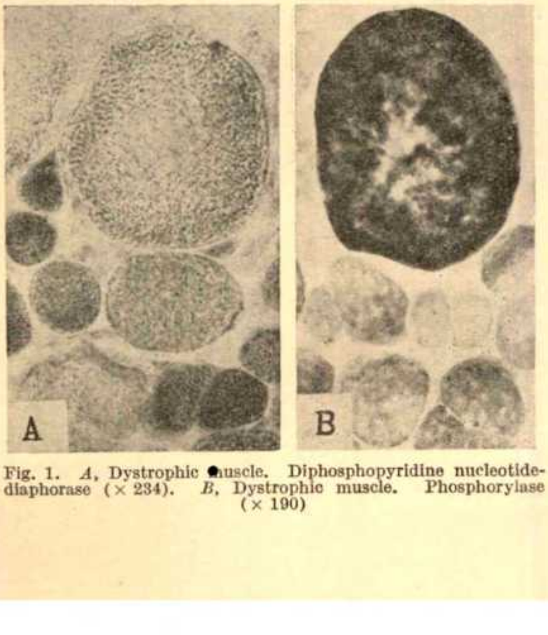

Fig. 1. *A*, Dystrophic muscle. Diphosphopyridine nucleotide-diaphorase (× 234). *B*, Dystrophic muscle. Phosphorylase (× 190)

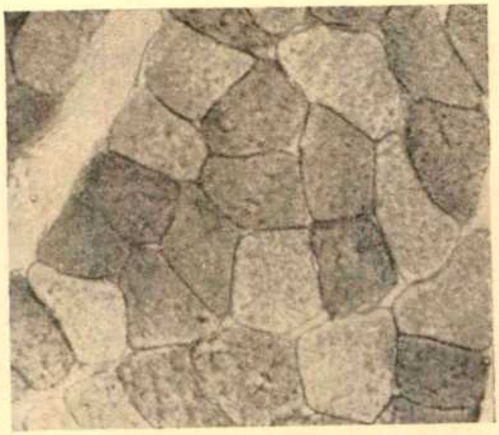

Fig. 2. Normal human tibialis anterior. Lactate dehydrogenase (× 208)

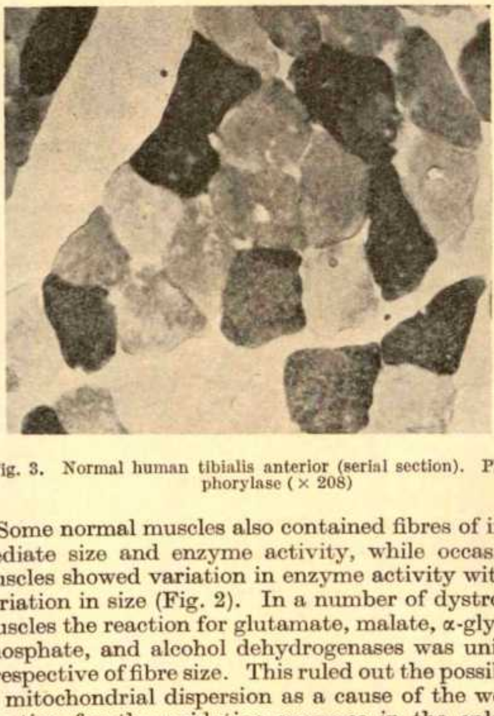

Fig. 3. Normal human tibialis anterior (serial section). Phosphorylase (× 208)

Some normal muscles also contained fibres of intermediate size and enzyme activity, while occasional muscles showed variation in enzyme activity without variation in size (Fig. 2). In a number of dystrophic muscles the reaction for glutamate, malate, α-glycerophosphate, and alcohol dehydrogenases was uniform irrespective of fibre size. This ruled out the possibility of mitochondrial dispersion as a cause of the weaker reaction for the oxidative enzymes in the enlarged fibres.

It appears, therefore, that normal human muscle is composed of two types of fibre; one, generally of smaller diameter, is strong in intramitochondrial oxidative enzymes and weak in phosphorylase, while the other, usually larger, reacts conversely. The atrophic and hypertrophic fibres in muscular dystrophy correspond to these two types respectively. This observation may explain why in muscular dystrophy some fibres hypertrophy while others atrophy. Variations in the proportion of the two types of fibres may account for the earlier and more frequent involvement of certain muscle groups by the dystrophic process.

A reciprocal relationship between phosphorylase and oxidative enzyme activity with the corresponding variation in fibre size was also demonstrated in the muscle fibres of the rat and pigeon. Different muscles contained varying proportions of the fibre types.

The pectoralis muscle of the pigeon, a red muscle, consisted mainly of small fibres (20–35μ) rich in oxidative enzymes and poor in phosphorylase. It contained a few large fibres (40–50μ), with converse activity, located at the periphery of the muscle bundles.

In the toad and goldfish the small fibres were strong in oxidative enzymes as well as phosphorylase, while the large fibres were weak in both enzyme systems. All the muscles of the toad and the white muscles of the fish contained small, intermediate and large fibres. The red muscle of the fish is composed entirely of strongly reactive small fibres.

Assuming that phosphorylase reflects the ability to utilize and synthesize glycogen, it seems that in the larger muscle fibres in warm-blooded animals intrinsic glycogen is the main source of energy. The smaller fibres preferentially use the various substrates of the Krebs cycle. The high lactate dehydrogenase activity in muscle suggests that not all lactic acid produced in muscle is transported to the liver for further oxidation.

In the case of phosphorylase it is known that the inactive precursor is activated during contraction and by such procedures as freezing. The possibility therefore exists that one type of fibre may be preferentially activated.

On the other hand, the reciprocal enzyme activities may represent different phases in a cycle of activity associated with different states of contraction. This would explain the occurrence of fibres of intermediate size and activity. It seems impossible, however, for the hypertrophic and atrophic fibres in dystrophic muscle to be merely different phases of activity of the same fibre type.

The muscle of the fish and toad differs from the muscle of warm-blooded animals in that some fibres are strong in oxidative enzymes as well as phosphorylase, while others are weak in both. The marked variation in diameter between the fibres of different enzyme activity supports the view that these fibres are of different types, as does the consistency of both reaction and size in the red muscle of the fish.

The small muscle fibres with strong oxidative enzyme and weak phosphorylase activity seem to correspond to the red muscle fibres in animals. There is no apparent correlation of fibre types with the fast and slow fibres of amphibia.

A detailed report of this work will be published elsewhere.

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[1] Wachstein, M., and Meisel, E., *J. Biophys. Biochem. Cytol.*, 1, 483 (1955).

[2] Nachmias, V. T., and Padykula, H. A., *J. Biophys. Biochem. Cytol.*, 4, 47 (1958).

[3] Ogata, T., *Acta Med. Okayama*, 12, 216 (1958).

[4] Beckett, E. B., and Bourne, G. H., *Acta Anat.*, 33, 289 (1958).

[5] Scarpelli, D. G., Hess, R., and Pearse, A. G. E., *J. Biophys. Biochem. Cytol.*, 4, 747 (1958).

[6] Hess, R., Scarpelli, D. G., and Pearse, A. G. E., *J. Biophys. Biochem. Cytol.*, 4, 753 (1958).

[7] Pearse, A. G. E., *J. Histochem. and Cytochem.*, 5, 515 (1957).

[8] Burstone, M. S., *J. Histochem. and Cytochem.*, 7, 112 (1959).

[9] Takeuchi, T., *J. Histochem. and Cytochem.*, 4, 84 (1956).

Mitotic Pattern and Histogenesis of the Central Nervous System

ROUNDED cells which repeat mitosis adding new cells to the tissue are found at the free margin of developing organs, both in plant and animal. In plants the germinal cells form cambium, while in various animal organs the germinal cells are arranged in an epithelium-like layer at the free margin of the tissues[1].

A typical example is observed in the developing central nervous system or the neural tube (Fig. 1). The germinal layer of the neural tube is called germinal epithelium. The cells in this germinal epithelium are supposed to produce new cells which constitute the mantle layer[2]. Close examination of these cells in the germinal epithelium reveals that: (1) the germinal cells are rounded and always found directly beneath the internal limiting membrane; (2) all the nuclei of the germinal cells show mitotic changes from pro-metaphase to early telophase; (3) their cytoplasm appears clear in a usual stain; (4) the direction of the mitosis is always parallel with the internal limiting membrane.

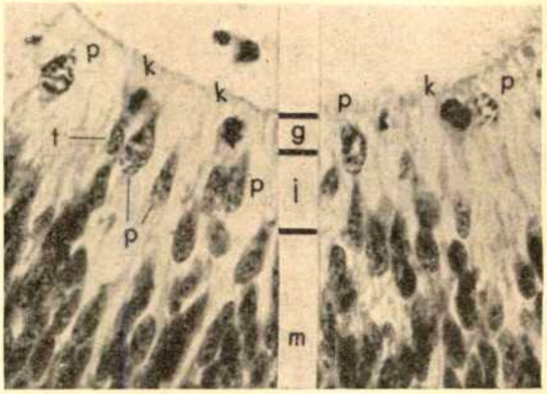

Fig. 1. The wall of the neural tube of the human embryo (C.R., 12 mm.). *g*, Germinal epithelium; *i*, intermediate zone; *m*, mantle layer; *k*, so-called germinal cell showing mitotic nucleus; *p*, prophase nucleus; *t*, nucleus of late telophase

Where an intermediate zone with sparse nuclei exists between the germinal epithelium and the mantle layer (Fig. 1), no resting cell is found in the germinal epithelium; whereas the resting cells are packed densely in the mantle layer. In the intermediate zone between the germinal epithelium and the mantle layer, nuclei at the latter half of telophase and early prophase are to be found. The prophase nucleus is swollen and enlarged, and the telophase nucleus is smaller[3]; but these nuclei in the intermediate zone show smooth transition to the resting cells in the mantle layer on one hand and to the mitotic germinal cells in the germinal epithelium on the other.

These observations can be explained if we assume that the following changes (Fig. 2) occur in sequence as the cells undergo mitosis. The resting cells in the mantle layer rise toward the internal limiting membrane as the cell begins to divide. (The emigration of the germinal cells to the internal limiting membrane during mitosis is considered to be associated with the cytoplasmic changes which round off[4] the mitotic cells, releasing them from the adjacent cells.) The cytoplasm of the cells becomes rounded and takes up less stain than usual. (The rounding off

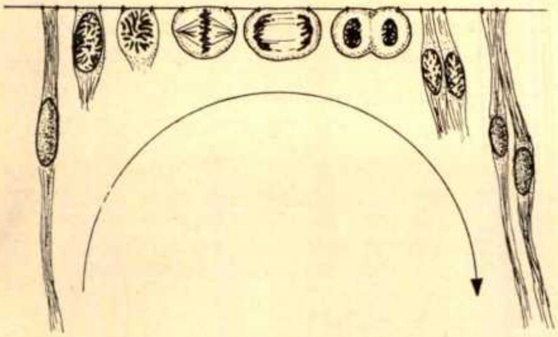

Fig. 2

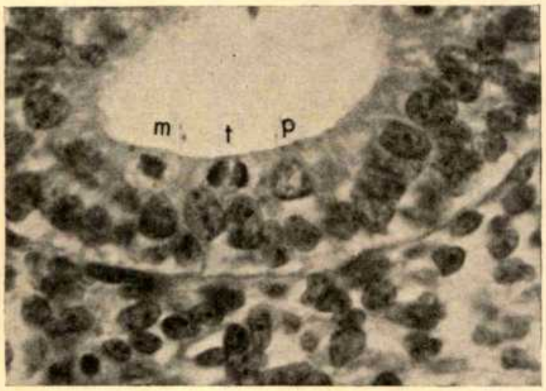

Fig. 3. Mitotic figures in the intestine of the same embryo as Fig. 1. *p*, Prophase nucleus; *m*, metaphase nucleus; *t*, telophase nucleus

of the mitotic cells seems to be related to the solution of the cytoplasm[5,6] and to insufficient supply of adenosine triphosphate to the contractile system in the cytoplasm during mitosis[7].) The chromosomes separate in the direction parallel with the internal limiting membrane, and, when the cells have passed through mitosis, they move back to the mantle layer.

Though this interpretation of cell migration has already been suggested by Sauer[8], his claim and the presence of the same mitotic pattern in a great variety of organs have never attracted attention. I have found this same mitotic pattern (as mentioned in (1)–(4) above) in the following tissues: *Adults*: the epithelium of the efferent duct of the epididymis; the gastric mucosa; the intestinal mucosa; the epithelium of the endometrial gland. *Embryo*: the epithelium of the branchial clefts; the tracheal and lung anlage; the gastric mucosa; the intestinal epithelium (Fig. 3); the epithelium of the bile duct system including the gall-bladder; the epithelium of the pancreas (these six tissues are of endodermal origin); the mesonephric tubule; the mesonephric duct; the metanephric tubule; thickened part of the mesothelium (these four are of mesodermal origin); the epithelium of the nasal pit; Rathke's cleft; the anlage of the retina; the neural tube (these four are of ectodermal origin). The same mitotic pattern is found without distinction of germ layer origin. What is common in these tissues is the fact that they consist of a single layer of cylindrical epithelium. Though the neural tube seems to be many-layered, it can be clearly demonstrated, using Golgi's silver method, that the wall of the tube is actually a modified single-layered epithelium (Fig. 4). Therefore, we conclude that the so-called germinal cells in these epithelial tissues including the neural tube are, in fact, not cells *sui generis* but the actual cells of the epithelium in mitotic phase.

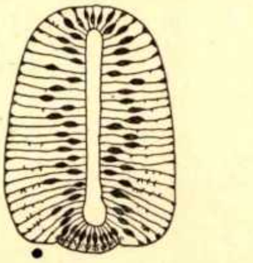

Fig. 4. Cord of chick embryo of the third day, stained by Golgi's method (after Cajal)

Concerning the development of the neural tube, this conclusion introduces a new point of view to the concept of the early histogenesis of the central nervous system, namely, the resting cells in the mantle layer that have been called ependymoblasts and primitive spongioblasts are nothing but the 'germinal cells' in the resting state. This concept, it is believed, contributes to the clearer understanding of the problem of the histogenesis of neurectodermal tumours[9] and of the regeneration of the central nervous system[10].

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[1] Swann, M. M., *Cancer Res.*, **181**, 1118 (1958).

[2] Ziehen, T., "Hertwig's Hdb. d. vergl. exp. Entw.-gesch. d. Wirbeltieren", **3**, 3 (Jena, 1906).

[3] Brachet, J., "Biochemical Cytology" (1957).

[4] Ris, H., in "Analysis of Development" (Saunders, Phil., 1955).

[5] Marsland, D., in "International Review of Cytology", **5** (Academic Press, New York, 1956).

[6] Heilbrun, L. V., "An Outline of General Physiology" (Saunders, Phil., 1952).

[7] Lettré, H., *Naturwiss.*, **38**, 490 (1951).

[8] Sauer, F. C., *J. Comp. Neurol.*, **62**, 377 (1935).

[9] Fujita, S., *Acta Path., Jap.* (in the press).

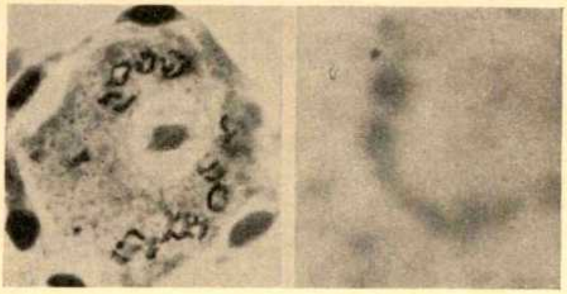
[10] Windle, W. F., "Regeneration on the Central Nervous System" (Charles C. Thomas, Springfield, U.S.A., 1955).

BIOLOGY

Electron Microscopy of the Golgi Apparatus

It has become customary in studies of the nerve cell by electron microscopy to apply the term 'Golgi apparatus' to appearances suggesting membranes, granules, or vacuoles[1]. In size, general configuration and osmiophilia the objects so designated cannot readily be recognized as forming part of the large intracellular 'network' familiar to workers with the light microscope. Moreover, adequate controls of these observations are prevented by the short periods (2–12 hr.) of osmication usual in the preparation of tissues for electron microscopy. As is well known, tissues must remain in osmic solutions for days or weeks before the capricious impregnation of the Golgi apparatus results. Further, it has been suggested that the initial buffering of the osmium fixative used in electron microscopy prevents for some reason the classical 'black reaction' of Kopsch (Gatenby, J. B., personal communication, 1958).

After experimentation, this difficulty has been circumvented and it has been found possible to view



(a) (b)

Fig. 1. *a*, Golgi network in light microscope photograph of a nerve cell (control). Palade fixation with post-osmication 30 days (× 1,200); *b*, portion of the Golgi net (thin section) photographed with the light microscope and further enlarged to × 10,000 for comparison with Fig. 2

with the electron microscope nerve tissues, known from light microscope controls to have undergone the necessary Kopsch reaction. Considerably lengthened schedules of osmication (30 days) followed by soaking in distilled water at 37° C. resulted in adequate impregnation of the apparatus in the controls, while preserving satisfactory fixation of the cytoplasm at the level of resolution of the electron microscope.

Fig. 1*a* is a photomicrograph at (× 1,200) of a dorsal root ganglion cell of a kitten that was fixed in buffered osmium (Palade fluid), and after post-osmication was embedded in wax and cut at 7μ. The cytoplasm is filled with the blackened networks described by Golgi[2]. Fig. 1*b* is from a 2μ section from the same block. In thin sections the network appears as individual thread-like bodies characteristically curved and beaded. One of these bodies photographed with the light microscope is enlarged to × 10,000 for comparison with Fig. 2.

Identically treated pieces of ganglion were embedded in methacrylate and cut to silver/gold sections with the ultramicrotome. On examination with the electron microscope, portions of the Golgi apparatus exactly comparable with those of Fig. 1*b* were easily recognizable during scanning and were selected for micrography (Fig. 2). From an examination of the electron micrographs certain inferences can be drawn.

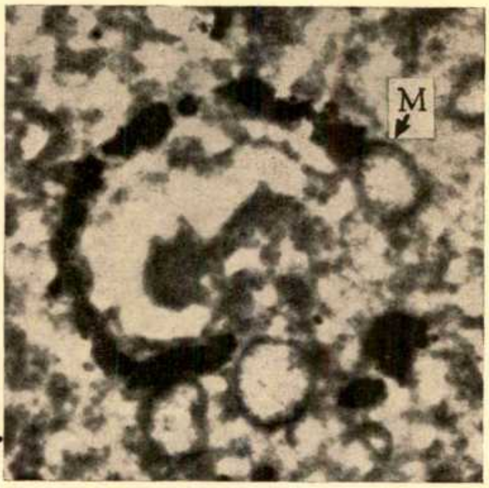


Fig. 2. Electron micrograph (× 22,000) showing Golgi net in close relation to vacuolated mitochondria, *M*

(1) The mitochondria (Fig. 2,*M*) are vacuolated in the kitten and are identical with those recorded by Barton and Causey[3] in ganglion cells fixed for only 2 hr. in buffered osmium.

(2) The mitochondria may be solitary or arranged in aggregates in close apposition like a chain of streptococci.

(3) Segments of the classical Golgi apparatus are clearly recognizable. They appear as a relatively coarse deposition of osmium particles.

(4) This deposition of osmium is unrelated to other 'preformed' structures in the cytoplasm such as the agranular reticulum or the acrosome-like piles of 'parallel plates' described by some workers[1] as the Golgi apparatus in the nerve cell.

(5) A clear space suggesting a shrinkage artefact may accompany the strands of osmium. It does not have a counterpart in the control material.

Repeated observation by the phase microscope on living ganglion cells has consistently failed to visualize any object comparable with the blackened bodies seen in the fixed cells (Figs. 1*a* and *b*, 2). Individual granules or aggregates of granules, comparable with the mitochondria of fixed cells, can however be readily seen. These findings will be discussed and a more extensive publication of the micrographs will appear elsewhere.

I wish to thank Dr. H. D. Purves for assistance and the Medical Research Council (N.Z.) for facilities.

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[1] Gatenby, J. B., and Lufty, R. G., *Nature*, **177**, 1027 (1956).
[2] Golgi, C., *Arch. Ital. Biol.*, **30**, 60 (1898).
[3] Barton, A. A., and Causey, G., *J. Anat.*, **92**, 399 (1958).

Abbreviated Development in a Pinnotherid Crab

ATKINS[1] has described the post-embryonic development of two British species of *Pinnotheres*. In *P. pisum* there are four zooeal stages and one megalopa, and in *P. pinnotheres* there are two zooeæ and a megalopa. In *P. pisum* the megalopa stage was reached six weeks after spawning and in *P. pinnotheres* it was reached after an interval of 21–26 days. Atkins quotes Sandoz and Hopkins[2] and Hart[3] to the effect that *P. ostreum* takes 21 days from hatching to megalopa and *P. taylori* 28 days from hatching to young crab.

P. moseri Rathbun is a tropical crab found in the atrial cavity of a large simple ascidian, *Ascidia nigra*. Recent observations show that the post-embryonic development of this crab is unusually rapid. Two berried females brought into the laboratory on October 21, 1959, liberated zooeæ during the nights of October 28 and 30, respectively. In both cases the zooeæ correspond closely to the fourth zooeal stage of *P. pisum* as described by Atkins, with the exception that the rostral spine is short, and lateral spines are absent. There are four swimming setæ on the maxillipeds and the thoracic limbs are well developed, the first pereiopod being chelate: in living specimens the thoracic limbs tend to be hidden under the carapace (Fig. 1).

The first batch of zooeæ moulted to megalopas in the early morning of October 30, giving a maximum

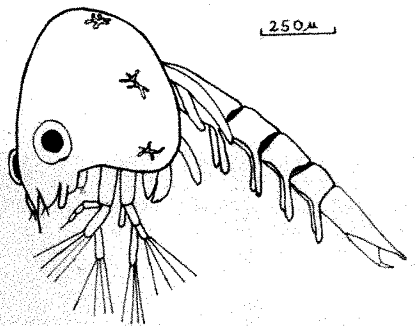

Fig. 1. *Pinnotheres moseri*, newly hatched zooea drawn from a living specimen with the abdomen extended. Four of the thoracic appendages are visible and also portions of the right maxillipeds appearing below those of the left side

time between hatching and attainment of megalopa of 36 hr. In the second batch of larvæ moulting to the megalopa was first seen at 17.30 hr. on October 31, thus giving a maximum of about 24 hr. from hatching to megalopa. As both batches hatched during the night the exact times are not known. The larvæ in both cases were kept at about 29° C., which was within 1° C. of sea water temperatures in October.

The post-embryonic development of *P. moseri* is thus considerably abbreviated when compared with what is known of other species of Pinnotheres. The significance of such abbreviation cannot at present be assessed; but it is pertinent to point out that *Ascidia nigra* is confined in its distribution to sheltered water such as enclosed bays and lagoons and is absent from communities of animals in more open areas. If *P. moseri* is an obligate commensal in *A. nigra* and cannot live in other hosts, an abbreviated larval development might be advantageous in preventing the larvæ from dispersing too far away from the concentrations of its host population.

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[1] Atkins, D., *Proc. Zoo. Soc. Lond.*, **124** (4), 687 (1955).

[2] Sandoz, M., and Hopkins, S. H., *Biol. Bull., Woods Hole*, **93**, 250 (1947).

[3] Hart, J. F. L., *Canad. J. Res.*, **12**, 411 (1935).

Calcium–Lithium Competition in Absorption by Plant Roots

In the process of absorption by plant roots, different ions may compete with each other, may fail to compete, or finally, one kind of ion may increase the rate of absorption of another. These specificities are considered due to the existence of specific binding sites on carriers which transport the ions across cellular membranes. Examples of mutually competing ions are the pairs potassium–rubidium[1] and calcium–strontium[2]. Neither sodium nor lithium competes readily for the potassium–rubidium sites, at low to moderate concentrations, and magnesium is bound loosely if at all by the sites which transport calcium and strontium.

In work with plants, all reported cases of specific mutual competition among mineral ions have involved pairs of ions closely related chemically. However, calcium severely inhibits the absorption of lithium by barley roots (ref. 3 and Epstein, E., unpublished work), and it has now been determined that the interference is competitive, indicating identical binding sites for these two ions.

Barley, variety Arivat, was grown in dilute calcium sulphate as described earlier[2]. The absorption experiments were conducted with 1-gm. portions of excised roots in 100 ml. volumes of aerated solutions at 30° C., for 60 min., during which absorption was a strictly linear function of time. The concentration of lithium chloride was varied from 1·0 to 10·0 m.moles/l., in the absence and presence of calcium sulphate at 1×10^{-4} and 1×10^{-3} *M*. The roots were rinsed, ashed at 500° C., and lithium in a solution of the ash was determined by flame spectrophotometry at 671 mμ. In Fig. 1, the reciprocal of the rate of absorption, v (in μmoles/gm./hr.), is plotted against the reciprocal of the lithium concentration, S (in m.moles/l.). In the presence of calcium, the slope of the line increases without change of the intercept, indicating competitive interference by calcium with lithium absorption.

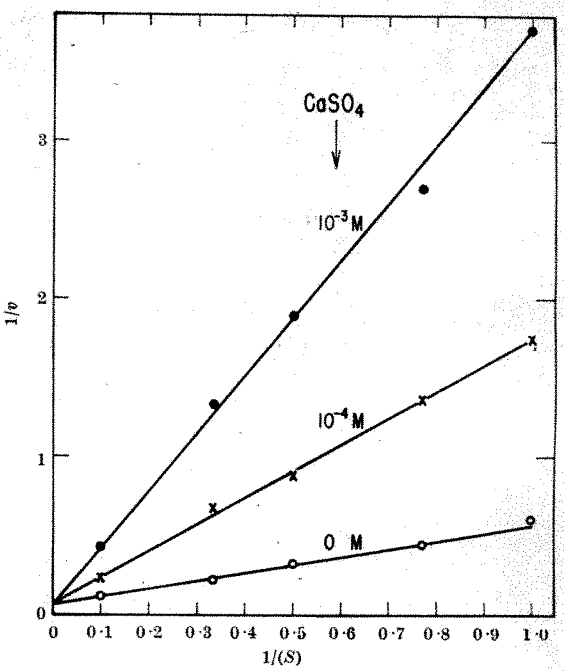

Fig. 1. Competitive inhibition of lithium absorption by calcium

Calcium, which is here shown to inhibit severely and competitively the absorption of lithium, accelerates the absorption of several other ions, among them potassium[4] and rubidium (Table 1). These findings confirm the earlier conclusion[1], recently questioned by Sutcliffe[5], that there are sites transporting lithium which are distinct from others which transport potassium and rubidium, calcium strongly competing with lithium for the former sites while increasing the rate of potassium–rubidium transport.

Table 1. Effect of Calcium on Absorption of Rubidium by Barley Roots

$CaCl_2$, *M* × 10^3	0	1	2	5	10	25
Rb* uptake, μmole/gm. fresh wt./hr.	8·9	11·8	13·3	14·8	16·0	16·5
$CaSO_4$, *M* × 10^3	0	1	2	5	10	
Rb* uptake, μmole/gm. fresh wt./hr.	8·9	9·8	11·1	11·0	11·8	

* Rubidium chloride, 1×10^{-3} *M*. Rubidium labelled with rubidium-86, assayed by determination of the radioactivity of the root ash. Experimental conditions and procedures as described in the text for the experiment on $Ca^{++} - Li^{+}$ competition.

At first it seems surprising to find competition between two such dissimilar ions as an alkaline earth cation, calcium, and an alkali cation, lithium. However, it is recognized that the small size and relatively large ionizing potential of lithium cause some resemblance between it and the alkaline earth cations[6]. An instance illustrating this resemblance is the low solubility of the carbonate and phosphate of lithium. Another example concerns the expansion of mica type crystal lattices as it depends on the interlayer cations. Barshad[7] has shown that montmorillonite saturated with lithium is similar in swelling behaviour and differential thermal analysis to samples with alkaline earth interlayer cations, and differs characteristically from samples saturated with the other alkali cations or ammonium ions. Finally, lithium, like calcium, increases the rate of rubidium absorption by barley roots[1].

These similarities seem to reflect ionic properties important in determining the 'fit' of the ions in molecular groupings of ligands. Lithium ions resemble the alkaline earth cations more than the other alkali cations in some of the properties that determine chemical affinities, including affinity for the carrier sites involved in active ion transport.

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[1] Epstein, E., and Hagen, C. E., *Plant Physiol.*, **27**, 457 (1952).

[2] Epstein, E., and Leggett, J. E., *Amer. J. Bot.*, **41**, 785 (1954).

[3] Jacobson, L., Moore, D. P., and Hannapel, R. J., *Plant Physiol.* (in the press).

[4] Overstreet, R., Jacobson, L., and Handley, R., *Plant Physiol.*, **27**, 583 (1952).

[5] Sutcliffe, J. F., *Biol. Rev.*, **34**, 159 (1959).

[6] Latimer, W. M., and Hildebrand, J. H., "Reference Book of Inorganic Chemistry", third ed. (The Macmillan Co., New York, 1951).

[7] Barshad, I., *Amer. Mineralogist*, **35**, 225 (1950).

RADIOBIOLOGY

Resistance of Algae to Radiation

Experiments carried out since 1954 have shown that various algae are much more resistant to radiation than are higher plants and animals. The results, which will be published fully elsewhere, are as follows:

Algae	Author	Dose survived (rads)
Spirogyra	Godward, 1954, and unpub.	20,000 plus
Zygnema, Mougeotia	Prasad, 1959, unpub.	50,000–70,000
Cosmarium	Nizam, 1959, unpub.	70,000
Euglena	Leedale, 1958	150,000
Eudorina	Rayns, 1959, unpub.	100,000
Chlorella	Nizam, 1959, unpub.	two successive doses of 1,000,000
Chaetomorpha	Patel, 1959, unpub.	50,000 plus
Prorocentrum	Dodge, 1959, unpub.	2,000

Of the above, only the last-mentioned, *Prorocentrum*, has the same order of sensitivity as the higher plants, if one excepts their specially resistant parts such as pollen grains and seeds. In all the experiments the algae were maintained in culture as long as possible after radiation; the recovery could follow in a matter of days but might need up to five weeks. Special features of the nuclear cytology probably account for the resistance and rapid recovery of the Conjugales[2,3,5] and *Euglena*[4]. These features do not occur in *Eudorina*, which has, however, very small chromosomes (ref. 1 and Rayns, unpublished work). Smaller still and unresolvable with the visual light microscope except at prophase, and even then uncountable, are those of *Chlorella*, which it seems impossible to kill. On the other hand, the chromosomes of *Prorocentrum* are relatively large, and it would seem that, like those of the higher plants in general, they may have a localized centromere. In those algae where the chromosome organization is conventional or at any rate gross, it seems that resistance to radiation is relatively low. No organism has, to my knowledge, been reported as surviving an experimental dose such as that given above to *Chlorella*, where the recovery process was, however, long; although it seems clear that the marine algae which received considerable radiation from atomic explosions were not killed.

The radiations were carried out at the Medical Research Council Unit, Hammersmith Hospital.

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[2] Godward, M. B. E., Proc. Int. Bot. Congr., Paris, 1954; *Heredity*, **11** (1957).

[3] King, G. C., *New Phyt.* (in the press).

[4] Leedale, G. F., *Archiv Mikrobiol.*, **32**, 32 (1958).

[5] Prasad, B. N., thesis, Ph.D., London (1959).

Radiocarbon Dating of Soil Humus

The turn-over of soil organic matter is a much-discussed problem in pedology. Although much has been written about the rate of breakdown of soil organic matter in mull and mor (for references see ref. 1), our knowledge of the turn-over of forest soil humus is very incomplete. This is particularly true of the organic matter in soil horizons below the humus layer.

The radiocarbon dating method opens a way to determine the approximate 'time of residence' in the soil of the organic matter, supposing that this time is of the order some hundred years or more. For this reason three soil samples from a profile in Garpenberg, Province of Dalarna, Middle Sweden, have been studied. The sample plot was a good-growing 95-year old spruce forest on moraine, altitude 200 m. (just above the so-called highest coastline). The samples represented the mor layer (A_0), the leached but humus-coloured top horizon of the mineral soil (A_1), and the brown accumulation horizon (B). The profile, which will be described in more detail in a later publication, is a favourable type of the podzol profile; similar profiles are not uncommon on moraine soils in this region.

The samples were passed through a 3-mm. sieve leaving stones, gravel and all but the finest roots on the sieve. The sifting process passed 63 per cent of the A_0, 90 per cent of the A_1 and 76 per cent of the B layers, based on samples collected in the field (measured dry).

The loss on ignition was 45·5 per cent of the A_0 sample, 15·9 of the A_1 and 5·2 of the B sample. In A_0 and A_1 the loss on ignition can be considered as estimates of the organic matter content, which, however, is lower than 5·2 in the B horizon.

The residual carbon-14 activity was measured with the following results:

Tree rings of oak grown 1845–1855 in Stockholm. Corrected for decay up to 1959	100·0 ± 0·3 per cent
St. 472, Sample A_0	98·3 ± 0·5 ,,
St. 473, Sample A_1	98·0 ± 0·5 ,,
St. 474, Sample B	95·6 ± 0·5 ,,

The activities have been corrected to the same initial isotopic composition by measuring the ratio of carbon-13/carbon-12 in all four cases (see Östlund[2]).

Owing to the well-known decrease of carbon-14 activity in the biosphere between 1850 and 1954 caused by industrial fuel combustion (Suess-effect), it is not possible to draw any reliable conclusion about the time of residence for samples A_0 and A_1. Sample B, however, shows a decrease of activity which is larger than the Suess effect. The calculated value for the time of residence is 370 yr., but the experimental errors give a total time-span of ± 100 yr. (95 per cent probability). In addition, the variations of the carbon-14 assay of the biosphere in times also before 1850 (de Vries[3]) add an extra unknown uncertainty.

Considering the fact that the site sampled probably has been covered with forest for about 9,000 years, the relatively low age of the organic matter in the B horizon suggests that the organic matter in this horizon (or a considerable part of it) is continually broken down and re-synthesized from substances coming from above. This is important, because in this and many other forest soil profiles the amount of organic matter in the B horizon equals or exceeds that in the A horizon. Further investigations seem called for, both by radiocarbon measurements and by other methods.

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[1] Handley, W. R. C., Forestry Comm. Bull., No. 23, 1 (London, 1954).
[2] Östlund, H. G., *Arkiv Kemi*, **12**, No. 6, 69 (1957); see p. 74.
[3] De Vries, H. L., *Proc. K. Ned. Akad. Wetensch.*, B, **61**, 94 (1958).

Accumulation of Cæsium-137 by Plants grown in Simulated Pond, Wet Meadow and Irrigated Field Environments

PLANTS grown in nutrient culture[1,2] and aquatic environments[3] accumulate more cæsium-137 than terrestrial plants. The work cited indicates that this isotope should be more available from fall-out under one set of environmental conditions than another. Consequently, the availability of cæsium-137 to plants was examined under conditions simulating shallow ponds, wet meadows and irrigated fields.

The spotted Persicaria or lady's thumb (*Polygonum persicaria* L.), which is tolerant of wide moisture conditions, was transplanted from the field into 7,000 gm. Ephrata Sandy Loam soil[1] in 'Pyrex' battery jars. Stable cæsium was not added to the soil because increased uptake of cæsium-137 with addition of carrier cæsium has been shown[4]. After the plants were well established, 72·3 µc. of $^{137}CsCl_2$ in distilled water was added uniformly to the surface of water or soil, care being taken to avoid contamination of the aerial surfaces.

Plants were grown outdoors under conditions shown in Table 1. There were three pots for each environment and 12 plants per pot. Water-levels in the 'shallow pond' and 'wet meadow' cultures were maintained by adding distilled water with automatic devices while moisture in the 'irrigated field' cultures was maintained by replacing weight loss daily.

Table 1. CONCENTRATION OF CÆSIUM-137 IN PLANTS GROWN IN DIFFERENT ENVIRONMENTS

Environment simulated	Experimental condition	Mean value at 5 hr.* (µc./gm.)	Uptake ratio, field capacity = 1	Mean value at 36 days (µc./gm.)	Uptake ratio
Shallow pond	Saturated soil with 2½ in. standing water	$3·1\times10^{-3}$ ±0·86	75	$1·4\times10^{-2}$ ±0·32	450
Wet meadows	Saturated soil, water table ½ in.	$8·2\times10^{-5}$ ±1·40	2	$8·9\times10^{-4}$ ±0·92	30
Irrigated field	Soil moistened to field capacity	$4·2\times10^{-5}$ ±0·65	1	$3·1\times10^{-5}$ ±0·67	1

* Each mean represents nine samples.

Plant stem tips (apical bud, stem and first two leaves) were sampled at intervals over a 36-day period. Samples were wet-ashed in nitric acid at temperatures less than 500° C. Radioactivity of the samples was measured with an end-window Geiger–Müller tube. Results of sampling at 5 hr. and 36 days are shown in Table 1.

The uptake ratio for plants grown in shallow standing water, saturated and field-capacity soils was 450 : 30 : 1. The high uptake in standing water is in close agreement with results obtained with emergent plants in an aquatic community[3], and not appreciably different from nutrient solution studies[2].

The greater uptake of cæsium-137 by plants grown in soil covered by shallow water and saturated soils is due mainly to the shallow root systems produced by plants in such environments, although some absorption by submerged leaves and stems would be expected. These root systems lie close to the surface in saturated soils, and adventitious roots, as in this experiment, often invade the water proper[5-7]. Thus, cæsium-137 can be absorbed directly from the water before the ions are fixed on to the soil. This is supported by results (Table 1, column 3) which show 20 per cent of total accumulation occurred during the first 5 hr.

The radioactivity of plants grown in field-capacity moisture was not appreciably greater than the level in plants taken from the natural habitat. This confirms reports of low availability of cæsium-137 in drier soils[2].

When applied to fall-out, the results reported here may be of considerable interest. Because uptake via roots is additive to direct fall-out on to the aerial plant surfaces, animals grazing in areas in which a major portion of the forage is from wet-lands may ingest cæsium-137 to 400-fold or more over those grazing on dry-lands. Similarly, crops grown in saturated soils or standing water should contain more cæsium-137 than crops grown on drier soils. Thus, increased accumulation of cæsium-137 from wet-lands may explain deviations from a correlation between rainfall and milk content[8,9] and added root absorption may contribute to the high cæsium-137 content of rice in Japan[10]. Further, a few dairy herds, grazing wet or periodically flooded meadows and edges of shallow ponds, might contribute disproportionately to the cæsium-137 content of a local milk supply, and extensive wet-lands could elevate the contamination-level of a larger region as has been suggested by Russell[11] relative to strontium-90 content of milk in England.

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[1] Rediske, J. H., and Hungate, F. P., Proc. Internat. Conf. Peaceful Uses Atomic Energy, **13**, 354 (1955).

[2] Klechkovsky, V. M. (ed.), Acad. Sci. U.S.S.R. (*A.E.C.-Tr*-2867) (1957).

[3] Pendleton, R. C., and Hanson, W. C., Proc. Second Internat. Conf. Peaceful Uses Atomic Energy, **18**, 419 (1958).

[4] Fredriksson, L., Eriksson, B., Rasmuson, B., Gahne, B., Edvarson, K., and Löw, K., Proc. Second Internat. Conf. Peaceful Uses Atomic Energy, **18**, 449 (1958).

[5] Emerson, F., *Bot. Gaz.*, **72**, 359 (1921).

[6] Dean, B. E., *Plant Physiol.*, **8**, 203 (1933).

[7] Alberda, T., *Plant and Soil*, **5**, 1 (1953).

[8] Booker, D. V., *Nature*, **183**, 921 (1959).

[9] Anderson, Ernest C., *Science*, **128**, 882 (1958).

[10] U.N. Scientific Committee, Supp. No. 17 (*A*/3838), 108 (1958).

[11] Russell, R. Scott, *Nature*, **182**, 834 (1958).

ENTOMOLOGY

Automatic Time/Response Records with *Drosophila*

Drosophila melanogaster Meig. is widely used for bioassay of insecticides. In developing a method for bioassay of insecticide residues in soil, we have found it necessary to use a time/response technique similar in principle to that employed by Callaway, Dirnhuber and Wilson[1]. In comparison with the more usual dose/response techniques, the sensitivity[2] and also the useful range of the assay can be increased by this means. However, with a time/response technique, two main difficulties arise when attempts are made to obtain records at precise times in order to follow accurately a given programme of observations. The response of each group of flies takes a finite time to assess and so, when many groups are involved, discrepancies occur between the desired and the actual times of the counts. Unless allowances are made for these time discrepancies—a tedious process—errors are introduced which can be important when the intervals between the records are small. Furthermore, it is often necessary to record responses throughout a 24-hr. period, or even longer, and this creates an obvious practical problem which, in the past, must have discouraged the use of time/response techniques as routine methods.

A brief description is given below of an apparatus which we have devised to record automatically the response of *Drosophila* at any number of pre-arranged times. The apparatus has worked successfully in this laboratory for the past six months and a detailed description of it is being prepared for publication elsewhere.

Acetone is used to extract the insecticide from the soil. Aliquots of the extracts are applied to 38 × 10 mm. absorbent cotton rolls, and the solvent is allowed to evaporate. A dilute solution of a syrup is added to each roll to provide food for, and so attract, the flies. The treated rolls are suspended in special 130 × 25 mm. 'exposure tubes' and 30 male *Drosophila* are put into each tube. The top of each tube is closed with muslin, but the bottom of the tube tapers evenly towards a 6-mm. diameter exit hole. A ventilation hole, 10 mm. in diameter, is present in the side-wall 30 mm. from the bottom of the tube. Thirty of these tubes are mounted vertically in a rack. In the base of the rack there are countersunk 6-mm. diameter holes into which fit the tapered bottoms of the exposure tubes. These holes are normally kept closed by a sliding shutter which has a complementary set of holes and is fitted underneath the base of the rack. The shutter is linked mechanically to two solenoids which open and close it in response to signals from a special timing device. The rack carrying the tubes is mounted immediately over a 152-mm. diameter cylinder which is coated with a non-setting gum and powered by an internal coiled spring. This can be wound up by a crank arm attached to a stub-axle passing through the end-plate of the cylinder. A pin-escapement, fitted externally to the other end-plate, limits the movement of the cylinder and is also operated by a solenoid activated by a signal from the timer.

The time of each operation of the apparatus is determined by an accurately driven tape in which holes have been punched at intervals to correspond with the required time sequence. An electrical contact detects each hole as it passes and the three signals required to work the solenoids of the apparatus are initiated. There is first a short pulse to open the shutter, followed immediately by a second, slightly longer pulse to close it and then, after a delay of about 5 sec., a third pulse causes the cylinder escapement to be released.

The timer is started as soon as flies have been put into half of the exposure tubes, which takes about 2 min., and, as the flies become affected by the insecti

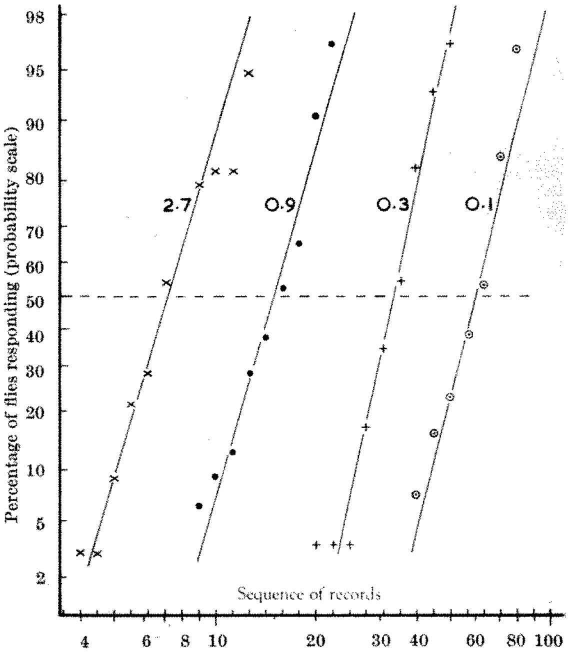

Fig. 1. Examples of time/response curves obtained with the recording apparatus for four standard doses of dieldrin. The figures refer to the dose of dieldrin in µgm. per roll. Temperature, 25 ± 0·5° C.; relative humidity, 72 ± 2 per cent

cide, they fall and collect in the recesses formed by the holes in the base of the rack. The apparatus is illuminated from above so that unaffected flies are attracted upwards and away from the exit holes. At an appropriate time, the shutter moves and opens the exit holes momentarily so that the affected flies fall out on to the sticky surface of the cylinder, where they are retained in discrete groups. The escapement is then released and allows the cylinder to move 1/16 of a revolution into position for the next record. At suitable times the number of flies in each position on the cylinder is noted, and the frequency results so obtained enable the time for 50 per cent response to be estimated in the usual way for each dose. The time/response curves shown in Fig. 1 are examples of results with this apparatus.

The programme of observations should be chosen so that there are equal grouping intervals for response to different doses and thus the programme used depends on the relationship between time of exposure and response to any given dose. Under our experimental conditions:

$$Y = a + b \log t$$

where Y is the probit of the percentage of flies responding to any given dose at time t, a is a function of dose in any one assay and b is the regression coefficient (Fig. 1). This relationship is that usually found when exposure to the poison is continuous[1,3,4] and it was thus necessary to arrange for observations to be made at equal intervals on a logarithmic, and not linear, time-scale; the tape was therefore programmed accordingly.

We wish to thank Mr. J. A. Nelder for numerous fruitful discussions while the apparatus was in the design stage, and also Mr. D. W. Wright for the help and encouragement which he has given throughout the work.

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[1] Callaway, S., Dirnhuber, P., and Wilson, K. M., *Nature*, **170**, 843 (1952).
[2] Bartlett, B. R., *J. Econ. Ent.*, **44**, 621 (1951).
[3] Bliss, C. I., *Ann. App. Biol.*, **24**, 815 (1937).
[4] Bliss, C. I., *Ann. Ent. Soc. Amer.*, **33**, 721 (1940).

Physiological Age of Codling Moth Females (*Cydia pomonella* (L.)) caught in Bait and Light Traps

SEVERAL recent papers have compared the effectiveness of bait and light traps as means of estimating changes in adult codling moth populations and of timing the application of sprays[1–3]. The comparison of these trapping devices was based, in each instance, on the total number of moths caught over periods considered to have been critical for the subsequent infestation of the crop. In these investigations no attention was paid to the quality of the respective catches beyond noting the difference in the relative frequency of females attracted to each type of trap.

When trap catches are used for recording moth activity as a guide to timing spray application, the age of females in samples may be of little importance if the weather conditions impose a fairly long developmental period on eggs. However, the physiological age of the trapped moths must be carefully considered when interpreting catch results obtained under warmer conditions. This point was emphasized by Nel[4], whose investigations showed that bait trap samples consist predominantly of spent, or half-spent, females.

Some additional information has been gained in the course of an experiment on the performance of bait and light traps carried out in an apple orchard near Canberra, Australia. Four bait traps and one light trap were operated daily in alternating positions from November 1958 until March 1959. A standard lure of fermenting molasses incorporating pine-tar oil[5] was used in the bait traps. The light source was an *HP* 125 W.B.C. mercury vapour lamp. Females obtained from both types of traps throughout the season were fixed and stained according to the method of Hamstead and Gould[6]. Each female was dissected, and an estimate of its physiological age made in strict conformity with Nel's techniques and criteria.

The age distribution of the females caught in bait traps at Canberra was found to be almost identical with that observed by Nel in South Africa. It is thus likely that most females are not attracted to the bait before some of their first-laid eggs have had time to hatch.

Table 1

	Physiological age I (young)	Physiological age II (middle-aged)	Physiological age III (old)	Total
Bait traps	14	39	36	89
Light trap	113	74	11	198

$\chi^2 = 70_{(2)}$ $P < 0{\cdot}001$

The picture is entirely different for the light trap, which drew samples of quite significantly younger females from the field population (Table 1). This feature may be added to the previously recognized advantages of light traps over bait pans in codling moth population studies.

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[1] Groves, J. R., Ann. Rep. East Malling Res. Sta., 1954, **146** (1955).
[2] Savary, A., and Baggiolini, M., *Ann. Agric. Suisse*, **56**, 827 (1955).
[3] Schneider, F., Vogel, W., and Wildbolz, T., *Schweiz. Z. Obst.- u. Weinb.*, **66**, 410 (1957).
[4] Nel, R. I., *Ent. Memoirs*, **2**, 55 (1940).
[5] Van Leeuwen, E. R., *J. Econ. Ent.*, **36**, 430 (1943).
[6] Hamstead, E. O., and Gould, E., *J. Econ. Ent.*, **43**, 724 (1950).

ANATOMY

A Highly Organized Structure within a Neurosecretory Vesicle

PREVIOUS electron microscope studies have demonstrated an abundance of vesicles 1000–3000 Å. in diameter in the terminal branches of neurosecretory axons in the neurohypophysis[1], and in the sinus glands and pericardial organs of crustaceans[2,3]. I have recently observed that some of these neurosecretory vesicles possess a highly organized internal structure.

The pericardial organs of the Mediterranean stomatopod *Squilla mantis* were prepared for electron microscope study by fixation either in buffered

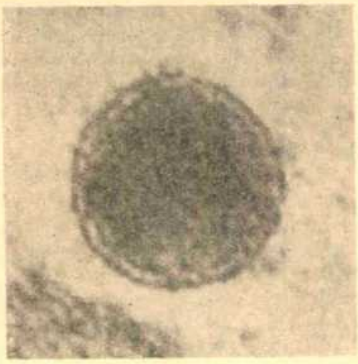

Fig. 1. A single neurosecretory vesicle of type *A* (× 225,000). Material fixed in buffered potassium permanganate, embedded in 'Araldite'

osmium tetroxide or buffered potassium permanganate and subsequent embedding in 'Vestopal *W*' or 'Araldite'.

In these preparations two types of vesicle could be clearly distinguished by their size and electron density and will be referred to here as types *A* and *B*.

Type *A* vesicles are regular in outline, spherical or slightly ovoid, approximately 1500 Å. in diameter. Each is surrounded by a distinct unit membrane, 75–90 Å. in thickness. The contents of type *A* vesicle can only be distinguished with difficulty as they are very electron dense. They appear to consist of tightly packed units without geometrical order, each unit comprising an electron-dense particle approximately 50 Å. in diameter, surrounded by a less dense sheath some 75 Å. in thickness (Fig. 1).

Type *B* vesicles, about 1200 Å. in diameter, are generally irregular in outline, though approximately spherical or ovoid; the bounding membrane is similar to that enveloping the type *A* vesicle, though less distinct, and there is usually a clear space between the membrane and the contents of the vesicle. The type of order displayed by the contents of the vesicle in various projections demonstrated in the electron micrographs is such as to suggest that the organization is based on a precise hexagonal packing of rod-shaped components. In the projection shown in Fig. 2 the structure appears as a series of alternating light and dark bands each measuring 60 Å. in width.

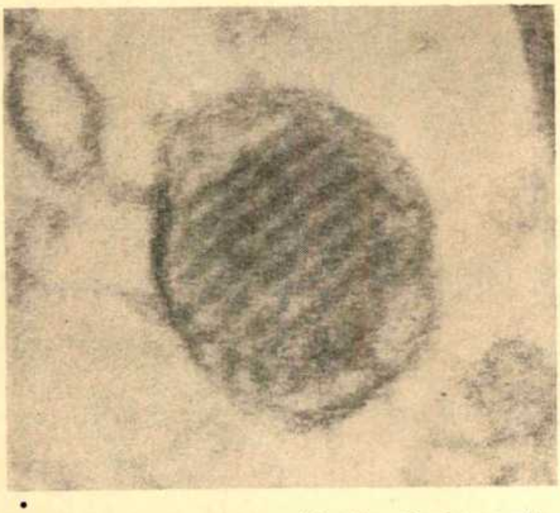

Fig. 2. A single neurosecretory vesicle of type *B* and two smaller vesicles (× 310,000). The orderly arrangement of material in the larger vesicle may be seen, and also a connecting link between the larger and one of the smaller vesicles. Material fixed in buffered osmium tetroxide, embedded in 'Vestopal *W*'

Fibres containing type *B* vesicles also contain smaller vesicles, spherical, ovoid or elongate in profile, approximately 500 Å. in diameter. There are indications that these small vesicles form part of a continuous tubular system in the axoplasm and that the type *B* vesicles are intimately related to this system. Indeed, it seems possible that the material in the type *B* vesicle may form within the system of small vesicles, and that the type *B* vesicles are swollen parts of this system within which material has assumed a crystal-like order.

The detection of orderly differentiated systems within neurosecretory particles is of theoretical interest, for there have been some indications from histochemistry also that neurosecretory vesicles may contain at least two chemical components[4]. The type of organization indicated by the present electron micrographs may, in fact, represent an orderly arrangement of these components which might permit the maximum of interaction between them in a suitable milieu.

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[1] Palay, S. L., *Anat. Rec.*, **121**, 348 (1955).
[2] Hodge, M. H., and Chapman, G. B., *J. Biophys. Biochem. Cytol.*, **4**, 571 (1958).
[3] Knowles, F. G. W., "Comparative Endocrinology", 223 (John Wiley, 1959).
[4] Hild, W., and Zetler, G., *Pflüg. Arch. ges. Physiol.*, **257**, 169 (1953).

'Synaptic Vesicles' in the Neurohypophysis

ELECTRON microscope studies of the neurohypophysis have shown that the terminal branches of the hypothalamo-hypophysial tract fibres contain, in addition to mitochondria, vesicles or granules of two kinds distinguished by their size[1,2]. The larger inclusions each have a diameter of the order of 100–300 mμ (1000–3000 Å.), and are thought to represent the secretory material which may be demonstrated by histochemical means, using the light microscope. The smaller inclusions, with diameters of approximately 30 mμ (300 Å.), have been described as synaptic vesicles[1] because they resemble in size inclusions found elsewhere in the nervous system at nerve terminals.

In the course of studying the neurohypophysis of rats and ferrets with the electron microscope, using material fixed either in buffered osmium tetroxide or in buffered potassium permanganate, we have observed a correlation of vesicle size with fibre size and fibre position. Small fibres, especially those closely applied to capillary walls, generally contain small vesicles, often densely packed and filling the whole outline of the fibre; in such fibres the larger vesicles are either absent or few in number (Figs. 1 and 2). In larger fibres the larger vesicles are found, either alone or with some small vesicles also. Large fibres containing small vesicles only have not been observed.

The correlation of vesicle size and fibre size is in itself not inconsistent with the view that the smaller vesicles are analogous to vesicles found at synaptic terminals. It is significant, however, that when large vesicles are found in fibres closely adjacent to capil

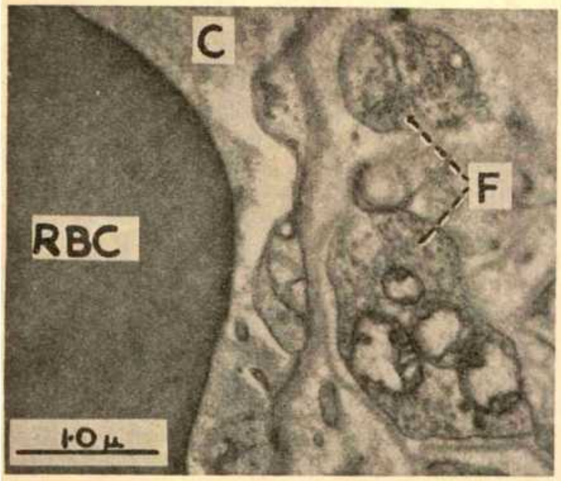

Fig. 1. A small portion of the neurohypophysis of a rat in which neurosecretory fibres (*F*) in close proximity to a blood capillary (*C*) are shown, and are seen to contain only small vesicles and mitochondria. *RBC*, part of a red blood cell. Fixed in buffered potassium permanganate, embedded in 'Araldite'

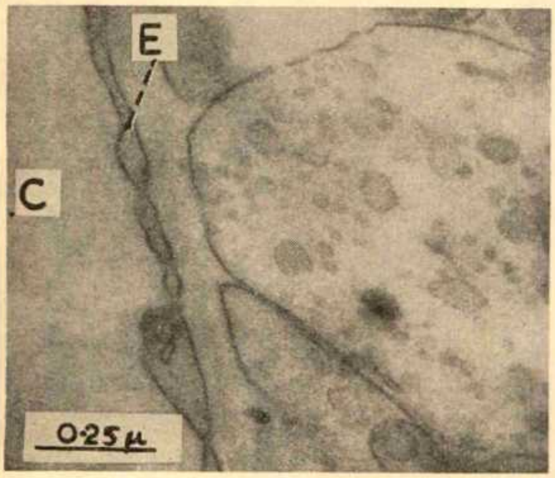

Fig. 2. A similar region in the neurohypophysis of a ferret. Indications of breakdown of large vesicles to small are apparent. *E*, fenestrated endothelium. Fixed in buffered potassium permanganate, embedded in 'Araldite'

aries, an internal structure may be detected in them which indicates that they may contain an aggregation of inclusions of the smaller type. There is, therefore, an interesting parallel between the vesicles in the nerve terminations of the mammalian neurohypophysis, and those in the pericardial organs of crustaceans, in which an orderly arrangement of small vesicles or particles within larger vesicles has been detected (previous communication).

As a result of these findings, we believe that it is imprudent at present to imply[1], by the use of the ame term, that all small vesicles in the terminations of neurosecretory fibres resemble functionally the ynaptic vesicles found elsewhere in the nervous ystem.

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Hartmann, J. F., *Z. Zellforsch.*, **48**, 291 (1958).

ARCHÆOLOGY

A New Palæolithic Find in Tapti Valley, Khandesh, India

In 1863 Foote reported a first palæolith from laterite gravels occurring at Pallavaram, near Madras. This find proved the presence of early man, maybe in the Pithecanthropoid or Neanderthaloid stage in India[1].

De Terra and Paterson[2] later recognized different phases of glacial cycle in Narbada valley. They were able to point out that early Palæolithic 'flake industries' were associated with Middle Pleistocene *Elephas antiqus* and *E. namadicus* and that three different cycles of sedimentation could be distinguished and correlated with the three stages in the formation of Narbada alluvium.

Pierre Teilhard de Chardin[3] of the Wenner–Gren Foundation, New York, points out in his good-will message to the Palæontological Society that "... first care should be taken to organize with the help of a few trained collectors, some kind of permanent surveyance: of the Narbada Basin (for the research of the hand-axes Man of India, skull or limb-bones...). Between S.E. Africa and Java, India happens to hold an exceptionally critical place, as far as the origin of Man is concerned".

During geological and ecological surveys of Khandesh Valley, I came across an interesting spot beside the Girna River bank near Central Railway bridge, 3 miles from Jalgaon, situated 21° 1′ N. and 75° 35′ E., 261 miles north-east of Bombay.

The River Girna rises in the hills west of Kalyan, in Nasik district. It is fed by streams from the northern slopes of the Sapta-Shring Range. After a course of about 150 miles, it joins the Tapti near Nander. Its course lies in equal parts in Nasik and Khandesh. Passing through Nasik almost in a straight line eastwards, its course changes to north-east in Khandesh. Near Jalgaon, it bends north and north-west, flowing for several miles with many windings almost parallel to the Tapti.

The underlying rock formation at places is Deccan trap. On the banks of the River Girna pebble conglomerate abound. They are capped by layers of sandy alluvium. This indicates that the river must have overflowed at times. The river now appears to be eroding ancient beds and underlying rocks.

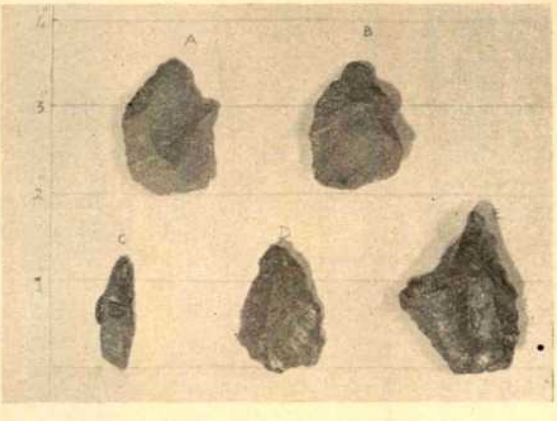

Fig. 1. Scrapers from the Girna River banks

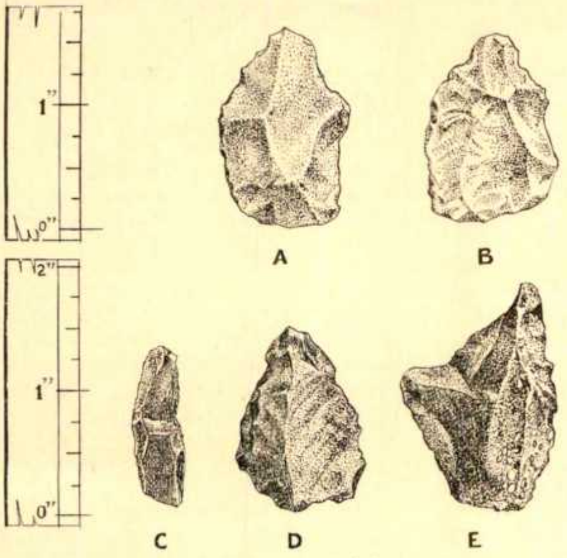

Fig. 2. Scrapers (dorsal view)

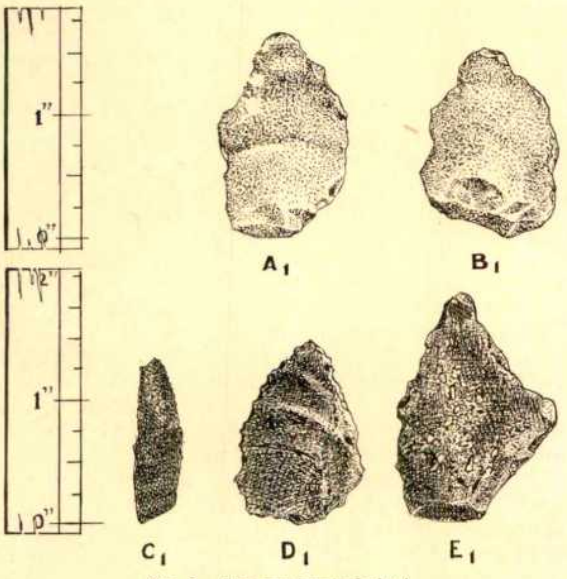

Fig. 3. Scrapers (ventral view)

The specimens of flakes could be of upper Acheulian tools belonging to early Sohan industries[2]. The scrapers show resemblances with early Soan flakes.

To my knowledge there is no record of such palæolithic find hitherto from Khandesh, this being the first find of such type from the Girna River in the Tapti Valley from Khandesh. It is suggested that a systematic survey of the river valleys which has hitherto been neglected be undertaken by archæologists, as it may one day show rich finds since this area was once a Narbada basin during the post-Eocene era.

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[1] Foote, J., *J. Palaeo. Soc. India*, **1** (1), 28 (1956).

[2] De Terra and Paterson, "Studies in the Ice Age in India and Associated Human Cultures" (Washington, 1939).

[3] De Terra and Paterson, *J. Palaeo. Soc. India*, **1** (1) (1956).

EDUCATION

" 'Teen-Age' Education in England"

You have, of course, a perfect right to say (*Natur* February 6, 1960) that, in your opinion, the Crowthe Report has made a wrong choice of priorities, bu have you the right to leave out of your article an reference to the conditions that that Report lay down as essential if the age is to be raised ? I calculates the number of extra teachers that will b needed not only to cover the extra year, but als at the same time to reduce over-large classes. I concludes that the task of providing these teacher though formidable, is not beyond the capacity o the country, if it is determined to accept the pla With regard to curriculum, it says that a great variety of courses must be provided, that method of teaching and discipline should be altered so tha the fifteens–sixteens will be treated more like adult and less like schoolchildren, and that staffing ratio should be generous enough to allow "for some adul leadership out of school hours".

Readers of the article in *Nature* who have no read the Report itself would not know that it contemplates these changes before the age can b raised.

With regard to the note by what is called a "stron minority of the Committee", which did not, incidentally, include any of the heads of secondary moder schools, this dealt with the treatment of a sma minority of children. The signatories go out of thei way to state : "To avoid any possibility of misunderstanding, we want to make it clear that thi note does not imply any dissent on our part fro the major recommendation that the school-leavin age should be raised. On the contrary, we feel confident that provision on the lines we have suggeste of a small minority would be held in public opinio to reaffirm the case for keeping the great majorit in school for another year. We certainly regard i ourselves." Apparently, they have not avoide misunderstanding on your part.

It is stated in *Nature* that "No member of th Crowther Committee teaches the lower streams o Modern Schools". This is equivalent to the charg that the three heads of secondary modern and on head of a comprehensive school are ignorant of th conditions which exist in their schools—a grav indictment indeed that would have to be supporte with strong evidence.

Since many States in the United States of Ameri have had a school-leaving age of sixteen for mo than twenty years, and some, one of 17+, wh should we assume that teachers in England are n able to make a sixteen leaving age "socially acceptable" if their counterparts across the Atlantic hav already done so ? Teachers in England have alway responded to the increasing demands that we hav made upon them, and I, for one, refuse to believ that they will fail the nation now, provided that w give them the right conditions for work.

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FORTHCOMING EVENTS

(*Meetings marked with an asterisk * are open to the public*)

Monday, March 7

University of London (at the Postgraduate Medical School of ndon, Ducane Road, London, W.12), at 4 p.m.—Dr. J. C. White . bnormal Hæmoglobins".*

Royal Geographical Society (at 1 Kensington Gore, London, W.7), at 5 p.m.—Mr A. T. A. Learmonth: "Medical Geography India and Pakistan"

British Society for the Philosophy of Science (in the Joint aff Common Room, University College, Gower Street, London, C.1), at 5 30 p.m.—Mr. Arthur Koestler: "Some Aspects of the ychology of Discovery".

Institution of Electrical Engineers, Electronics and Com- nications Section (at Savoy Place, London, W.C.2), at 5.30 p.m Dr P. N. Butcher: "An Introduction to the Theory of Masers h particular reference to the Travelling-Wave Maser".

Society of Engineers (at the Geological Society, Burlington use, Piccadilly, London, W.1), at 5 30 p.m.—Mr. R Lenoir . rogress of British High Grade Watch Production".

University of London (at King's College, Strand, London, W.C.2), 5 30 p.m —Dr. Bernard O. Phinney (University of California) he Use of Biochemical Genetics in the Study of Gibberellin-con- lled Growth".*

Society of Chemical Industry, London Section (at 14 Bel- ve Square, London, S.W.1), at 6.30 p.m.—Dr. W. Gerrard: "Recent lvances in the Chemistry and Industrial Applications of Hydrido- d Organo-Boron Compounds".

Society of Instrument Technology (at Manson House, 26 Port- ad Place, London, W 1), at 7 p.m —Mr. A. S. Younger, Mr. G. C. rgan and Mr. E. S. Mallett "Data Reduction for Guided Weapon ials at Aberporth".

Tuesday, March 8

University of London (in the Anatomy Theatre, University llege, Gower Street, London, W.C.1), at 1.15 p.m —Prof. W. B. aery: "A Middle Kingdon Fortress in the Sudan".*

University of London (at the London School of Hygiene and opical Medicine, Keppel Street, Gower Street, London, W.C.1), 5 15 p.m.—Dr. F. Sanger: "Amino Acid Sequences in Proteins".*

British Glaciological Society (at Bedford College, Regent's rk, London, N.W.1), at 5 30 p.m.—Annual General Meeting. Mr. Lister. "Drifting Snow".

Institute of Marine Engineers (at the Memorial Building, Mark Lane, London, E.C.3), at 5.30 p.m.—Cdr. R. G. J. Peaver, N.: "Distillation for Marine Purposes".

University of London (at Senate House, Malet Street, London, .C.1), at 5.30 p.m.—Prof. C. G. Hempel (Princeton University): ome Problems in the Logic of Probabilistic Explanation".*

Royal Aeronautical Society (at 4 Hamilton Place, London, .1), at 7 p.m.—Dr J. W. Granville: "Electronic Microcircuits".

Wednesday, March 9

Institute of Fuel (at the Institution of Civil Engineers, Great orge Street, London, S.W.1), at 5.30 p.m.—Mr. J. E. Richards . uclear Power for Marine Propulsion".

Institution of Electrical Engineers, Supply Section (at voy Place, London, W.C.2), at 5 30 p.m.—Dr. B. Salvage and Mr. A. M. Gibbons: "The Impulse Strength of Impregnated-Paper electrics as used in High-Voltage Cables".

University of London (at King's College, Strand, London, W.C.2), 5.30 p.m.—Dr. Bernard O. Phinney (University of California) ibberellins as Native Growth Regulators of Flowering Plants".*

Institute of Information Scientists (at the College of Pre- ptors, 2 Bloomsbury Square, London, W.C.1), at 6.15 p.m.—Prof. lin Cherry: "The Nature of Human Communication".

Society of Chemical Industry. Food Group (joint meeting with e Association of Public Analysts, at 14 Belgrave Square, London, W.1), at 6.15 p m.—Scientific Papers.

Royal Aeronautical Society (at 4 Hamilton Place, London, 1), at 7.30 p.m.—Mr. W. J. Egginton: "The Hovercraft and Its evelopment".

Thursday, March 10

Royal Society (at Burlington House, Piccadilly, London, W.1), 4.30 p m.—Dr. A. L. Hales: "Bernard Price Institute of Geo- ysical Research".

Institution of Electrical Engineers, Utilization Section t Savoy Place, London, W.C.2), at 5.30 p.m.—Mr. J. P. McBreen: ome Considerations in the Application of Power Rectifiers and nvertors".

Physical Society, Acoustics Group (in the Physics Department, perial College, Imperial Institute Road, South Kensington, Lon- n, S.W.7), at 5.30 p.m.—Dr. J. M. Bowsher: "Electronic Music".

University of London (at Senate House, University of London, ndon, W.C.1), at 5 30 p m.—Prof A. Hanau (University of Gott- gen): "Some Results of German Research in Agricultural Marketing d Prices".*

Chemical Society (in the Large Chemistry Lecture Theatre, Imperial College of Science and Technology, South Kensington, London, S.W.7), at 7.30 p.m —Prof. A. R. J. P. Ubbelohde, F.R.S.: "Ionic Crystals and their Melts" (Liversidge Lecture).

Friday, March 11

Royal Aeronautical Society (at the Institution of Mechanical Engineers, 1 Birdcage Walk, Westminster, London, S.W.1), at 9.45 a.m. —Symposium on "Vehicle and Instrumentation Problems in Upper Atmosphere Research". Speakers: Prof. Sir H. S. W. Massey, F.R.S., Mr. J. F. Hazell, Dr R. L F. Boyd, Dr. G. D. Robinson, Mr. H. J. Higgs and Mr. E. C. Cornford.

Physical Society, Low Temperature Group (at the Institute of Physics, 47 Belgrave Square, London, S.W.1), at 10 a.m.—Symposium on "Measurement Techniques (Flow, Pressure, Level, Temperature, etc.)".

Association of Applied Biologists (joint meeting with the British Ecological Society, at the British Museum (Natural History), South Kensington, London, S.W.7), at 10.50 a.m.—Meeting on "Ecological Effects of Crop Production".

Institute of Physics, Non-Destructive Testing Group (at 47 Belgrave Square, London, S.W.1), at 6 p.m.—Dr. R. Jones. "Analysis of Road Constructions".

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Dr. A. E. Werner: "Scientific Techniques in Art and Archæology".

Saturday, March 12

British Mycological Society (in the Botany Department, Birkbeck College, Malet Street, London, W.C.1), from 2.15 p.m. to 5 p.m — Conversazione.

London County Council (at the Horniman Museum, London Road, Forest Hill, London, S.E.23), at 3.30 p.m.—Miss A. G C. Grandison: "Collecting Reptiles and Amphibians in the United States of America".*

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned

Assistant Lecturer (with special qualifications or interests in radio astronomy) in Physics—The Registrar, The University, Manchester 13 (March 12).

Assistant Lecturer (with special interest in urban geography) in Geography—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1 (March 14).

Junior Lecturer (male or female, with an honours degree in chemistry or biochemistry, or A.R.I.C.) in the Department of Chemical Pathology—The Secretary, Westminster Medical School (University of London), Horseferry Road, London, S.W.1 (March 14).

Developmental Physiologist (with a good honours degree and preferably two or more years postgraduate research experience) in the Plant Physiology Unit, Department of Scientific and Industrial Research, Palmerston North, New Zealand, initially to assist in current studies using controlled climate facilities for investigation of the influence of the climatic factors on reproduction in plants of economic importance—The High Commissioner for New Zealand, 415 Strand, London, W.C.2, quoting Ref. No. B.11/21/26 and mentioning *Nature* (March 18).

Assistant Lecturer or Lecturer in Botany—The Secretary, The University, Exeter (March 19).

Imperial Chemical Industries Research Fellows in Chemistry, Physics, Engineering (including Chemical Engineering), Metallurgy, Pharmacology, or related subjects—The Registrar, The University, Sheffield 10 (March 19).

Lecturer in the Department of Sociology—The Registrar, The University, Leicester (March 19).

Research Fellow (engineer, physicist or mathematician) in Production Engineering—The Registrar, The University, Nottingham (March 19).

Lecturer or Assistant Lecturer (preferably with experience or interest in magnetic resonance) in Physics—The Registrar, University College of North Staffordshire, Keele, Staffs (March 21).

Lecturer or Assistant Lecturer (with a good honours degree) in Organic Chemistry—The Registrar, King's College (University of London), Strand, London, W.C.2 (March 21).

Research Assistant (graduate, with botany to the standard of a B.Sc. (General) degree) to the Head of the Department of Botany —The Secretary, Birkbeck College (University of London), Malet Street, London, W.C.1 (March 21).

Assistant Librarian (woman graduate)—The Principal, Royal Holloway College (University of London), Englefield Green, Surrey (March 22).

Demonstrator in the Department of Geology, to act as Curator of Collections—The Registrar, The University, Liverpool, quoting Ref. CV/T (March 25).

Lecturer in Agriculture (Animal Breeding)—The Registrar, King's College (University of Durham), Newcastle upon Tyne 1 (March 25).

Assistant Lecturer in Inorganic Chemistry; and Demonstrators (2) in Inorganic Chemistry—The Registrar, The University, Nottingham (March 26).

Assistant Lecturer in the Department of Chemistry—The Registrar, University College of South Wales and Monmouthshire, Cathays Park, Cardiff (March 26)

Chair of Metallurgy—The Registrar, University College of South Wales and Monmouthshire, Cathays Park, Cardiff (March 26)

Lecturer (with a good honours degree in geology and/or chemistry and research experience in geochemistry) in Geochemistry—The Secretary, The University, 38 North Bailey, Durham (March 26)

Lecturer (with an honours degree or equivalent) in the Department of Fluid Mechanics—The Registrar, The University, Liverpool, quoting Ref. CV/T (March 26).

Senior Scientific Officer (with a first- or second-class honours degree in science or engineering and at least three years postgraduate experience) at the Headquarters of the Department of Scientific and Industrial Research, London, S.W.1, for scientific administrative duties—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S W.1, quoting E.93/0/A (March 28).

Analytical Chemist (preferably with at least an honours degree with chemistry as a major subject, or a professional diploma of a recognized Chartered Institute) in the Government Analysts Laboratory, Salisbury, Federation of Rhodesia and Nyasaland—The Secretary (R), Rhodesia House, 429 Strand, London, W.C 2 (March 31).

Assistant Lecturer (with an honours degree in botany or agricultural botany, and preferably with research experience and an interest in plant physiology) in Agricultural Botany—The Registrar, The University, Leeds 2 (March 31).

Assistants (2) (with a good honours degree in geography and preferably with a special interest in physical geography and/or cartography) in the Department of Geography—The Secretary, The University, Edinburgh (March 31).

Lecturer in the Department of Pathology—The Secretary, The University, Edinburgh (March 31).

Plant Breeder and Geneticist in the Department of Agronomy (Plant Breeding Section), Waite Agricultural Research Institute, University of Adelaide, Australia, to undertake research on the factors affecting quality and yield in barley and to produce commercial barley varieties—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia, March 31).

Research Fellow (physicist, physical chemist or electrical engineer) in Gas Discharges, to work in association with Prof. G. Porter on an investigation of radiation from gas discharges—The Registrar, The University, Sheffield 10 (March 31).

Lecturer (preferably with special qualifications in engineering geology, including hydrology and soil science) in the Department of Geology, University of the Witwatersrand, Johannesburg, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa, April 15).

Lecturer (graduate in science) in Microbiology—The Secretary, The Queen's University, Belfast (April 30).

Research Fellow in the Department of Chemistry, for a research programme devoted mainly to investigating, in collaboration with Dr. W. P. Doyle, the ultra-violet absorption of thin films of inorganic solids—Prof. T. S. Wheeler, Department of Chemistry, University College, Dublin (May 1).

Assistant or Associate Professor of Mathematics—The Principal, Victoria College, Victoria, B.C., Canada.

Assistant or Associate Professor (with the degree of D Sc. or Ph.D. in zoology from a recognized university and special interests in the physiology of aquatic organisms, particularly fishes) in the Department of Zoology—The Chairman, Department of Zoology, University of Manitoba, Winnipeg 9, Manitoba, Canada.

Biochemist (Basic grade) in the Pathology Laboratory, to assist the Senior Biochemist—The Superintendent, Liverpool Royal Infirmary, Pembroke Place, Liverpool 1.

Regional Trials Officer (graduate in agriculture), for agricultural variety trials conducted by the National Institute of Agricultural Botany at the N.A.A.S. Experimental Husbandry Farm, Terrington St Clement, Norfolk; and an Assistant Trials Officer (preferably with practical experience in crop production), to work at Leeds University Farm, Headley Hall, Yorks—The Secretary, National Institute of Agricultural Botany, Huntingdon Road, Cambridge.

Scientific Officer/Senior Scientific Officer, Physical Chemist or Metallurgist (with a first- or second-class honours degree or equivalent) at the National Physical Laboratory, Teddington, Middlesex, for work in the Metallurgy Division on the use of radioactive isotopes as tracers in metallurgical investigations—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. F.648/9A.

Technician (preferably with some knowledge of physics, chemistry and biology, and experience in photography and section cutting) in the Electron Microscope Department—The Secretary, Strangeways Research Laboratory, Wort's Causeway, Cambridge.

REPORTS and other PUBLICATIONS

(*not included in the monthly Books Supplement*)

Great Britain and Ireland

The Society for Analytical Chemistry. Report of the Analytical Methods Committee, 1958. Pp. vi+24. (London: Society for Analytical Chemistry, 1959.) [2212

London County Council: Public Health Department—Scientific Branch. Annual Report of the Scientific Adviser for the year 1958. Pp. 42. (No. 4056.) (London: L.C.C., Public Health Department, 1959.) 1s. 3d. [2212

Imperial College of Science and Technology (University of London). Fifty-second Annual Report of the Governing Body, 1958–59. Pp. ii+102. (London: Imperial College of Science and Technology. University of London, 1959.) [2212

Royal Greenwich Observatory Bulletins. No. 16: Time Service 1959, April–June. Pp. 241–258. (London: H.M. Stationery Office, 1959.) 3s. 6d. net. [2212

Bowaters in North America. By Robert Sinclair. Pp. 48. (London: The Bowater Paper Corporation, Ltd., 1959.) [2212

Memoirs of the Geological Survey of Great Britain. Palæontology Vol. 4, Part 2: Fossil Plants of the Carboniferous Rocks of Grea Britain. By Dr. Robert Crookall. Second edition. Pp. i–lv+85–216+ plates 25–58 (London: H M. Stationery Office, 1959.) 110s. net. [221

Operator Training in Industry. By W. Douglas Seymour. Pp. 5 (London. Institute of Personnel Management, 1959.) 7s. 6d. [801

Other Countries

Population Reference Bureau, Inc., Washington, D.C. Populatio Bulletin, Vol. 15, No. 7: Japan's Population Miracle an Asia Solution to Asia's Greatest Problem. Pp. 117–136. (Washingto D.C.: Population Reference Bureau, Inc., 1959.) [801

Osmania University, Hyderabad. Publications of the Nizamia Observatory. Vol. 8, part 2: Comparison of the Places of Stars i the "Catalogue of 8824 Stars," Washington, with the Photograph Measures on the Plates of the Hyderabad Section of the Astrograph Catalogue. Under the direction of A. Ali. Pp. ii+27. Rs. 4; 6s. net Spectroheliographic Observations of Solar Flocculi made at th Nizamiah Observatory, Hyderabad, during 1951–1955, No. 1. B H. Alvi. Pp. 11. (Hyderabad: Osmania University Press, 1958 an 1959.) [301

Government of India: Ministry of Health Annual Report of th Central Drugs Laboratory from 1st April, 1955 to 31st March, 1956 Pp. i+46. Rs. 5.50; 8s. 6d. Report of the Central Drugs Laboratory 1st April, 1956 to 31st March, 1957. Pp. ii+47. Rs. 5.75; 9s. (Delhi Manager of Publications, 1959.) [301

Commonwealth of Australia Commonwealth Scientific and Industrial Research Organization. Annual Report—Division of Electrotechnology, 1958–59. Pp ii+21+9 plates. (Chippendale, N.S.W. Division of Electrotechnology, National Standards Laboratory 1959) [301

Population Reference Bureau, Inc., Washington, D.C. Population Bulletin, Vol. 15, No. 8: The New Awakening: Symposium of Officia Opinion. Pp. 137–160. (Washington, D.C. Population Referenc Bureau, Inc., 1959.) [301

Smithsonian Institution: Bureau of American Ethnology. Bulleti 143: Handbook of South American Indians, Vol. 7: Index. Pp vi+286. (Washington, D.C.: Government Printing Office, 1959 2 dollars. [301

National Science Foundation. Federal Funds for Science 8: Th Federal Research and Development Budget, Fiscal Years 1958 1959, and 1960. Pp. iii+74. (NSF-59-40.) (Washington, D.C. Government Printing Office, 1959.) 45 cents. [301

United States Department of the Interior. Fish and Wildli Service. North American Fauna No. 61 Fauna of the Aleutia Islands and Alaska Peninsula. By O. J. Murie. Invertebrates an Fishes collected in the Aleutians, 1936–38. By Victor B. Scheffer Pp. xiv+406. (Washington, D.C.: Government Printing Office 1959.) 1.25 dollars. [301

The Report of the President of the Carnegie Institution of Washington, 1958–1959. Pp. 54. (Reprinted from *Carnegie Institution o Washington Year Book* 58 for the year 1958–1959. Pages 1–54 (Washington, D.C.: Carnegie Institution of Washington, 1959.) [301

World Health Organization. Technical Report Series, No. 183 Mental Health Problems of Automation—Report of a Study Group Pp. 30. (Geneva: World Health Organization; London: H.M Stationery Office, 1959.) 1 Swiss franc; 1s. 9d, 0.30 dollars. [301

United States Department of the Interior: Fish and Wildlif Service. Fishery Bulletin 157: The Goldeye, *Amphiodon alosoide* (Rafinesque), in the Commercial Fishery of the Red Lakes, Minnesota By Marvin D. Grosslein and Lloyd L. Smith, Jr. Pp. iv+33–41. 1 cents. Fishery Bulletin 158: Food of the Squawfish *Ptychocheilu oregonensis* (Richardson) of the Lower Columbia River. By Richard B Thompson. Pp. iv+43–58. 20 cents. Fishery Bulletin 159: Effects o Fertilizing Bare Lake, Alaska, on Growth and Production of Red Salmon (*O. Nerka*) By Philip R. Nelson. Pp. iv+59–86. 25 cents Fishery Bulletin 160: Changes in Tuna Landings of the Hawaiia Longline Fishery, 1948–1956. By Richard S. Shomura. Pp. iv+87–106 20 cents (Washington, D.C.: Government Printing Office, 1959.) [301

British Territories in Borneo: Geological Survey Department Memoir No. 9: The Geology and Mineral Resources of the Sandaka Area and Parts of the Kinabatangan and Labuk Valleys, North Borneo By Dr. F. H. Fitch. Pp. xvi+202+40 plates. (Kuching, Sarawak Government Printing Office, 1958.) 6 Malayan dollars. [301

Transvaal Museum, Pretoria. Report for the year ending 31s March, 1959. Pp. ii+48. (Pretoria: Transvaal Museum, 1959) [301

Die Bildung Organischer Meterie aus Kohlensäure. Von Prof Dr. A. Eichinger. Studien uber Stärkebildung Ausserhalb der Pflanze Von Prof. Dr. A. Eichinger. (Bayreuth: Prof. Dr A. Eichinger Albert Preu-Strasse, 1. 1958 and 1959.) [301

West African Maize Research Unit. Trial Report, 1957. Pp. 40 (Ibadan. West African Maize Research Unit, Moor Plantation 1959.) [3012

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PROMOTION OF FUNDAMENTAL RESEARCH

IN recent reports, the National Science Foundation has consistently urged the strengthening of fundamental research, support of which, it has urged, is the key to the progress in science upon which technology depends. It has been widely recognized that the proportion of effort devoted to such research in the United States is inadequate, and the 488 million dollars allocated for that purpose in 1959 or the 494 million dollars in 1960 only represent some 6–7 per cent of the total allocation for research and development. Between 1956 and 1959, it is true, expenditure on fundamental research increased by 143 per cent, and now constitutes almost one third of the total research effort in the United States compared with about one fourth in 1956.

Dr. W. R. Brode, Science Adviser to the Secretary of State, in his presidential address to the American Association for the Advancement of Science on December 28, 1959, remarked on the recent increase in the support of fundamental research and the predicted doubling of such research during the next decade. This, he said, gives a high priority to questions of organization. It is of increasing importance to determine the most appropriate means of arranging for financial support, so as to avoid overlapping and ensure continuity ; equally, the determination of the right priorities, the selection of programmes and decisions as to their relative position, and the distribution of responsibility for their execution. Dr. Brode is obviously deeply concerned as to the way future policy in science is to be determined and executed in the American scene. Even with the vast resources at the disposal of the United States, neither man-power nor material resources are equal to all demands : selection is inevitable, and Dr. Brode is not satisfied that recent changes made by the President, such as the appointment of a Science Adviser and of a Science Advisory Committee, as well as of a Federal Council for Science and Technology, provide all that is required.

Dr. Brode is obviously watching with interest developments in Britain which may follow on Lord Hailsham's appointment as Minister for Science, and particularly the use which may be made of the Advisory Council for Scientific Policy, though he fully appreciates the wide difference between conditions in Great Britain and the United States. It is interesting, nevertheless, in view of the criticism already expressed in Parliament about the arrangements for representation of the Minister for Science in the House of Commons, to find that Dr. Brode is critical of the refusal of the chairman of the newly created Federal Council for Science and Technology to appear before a Congress Committee inquiring into the possible establishment of a Department of Science. The request was refused on the ground that the chairman was a privileged member of the President's staff. Dr. Brode suggests that while the President should continue to have a strong science adviser, it would be better to separate science co-ordination from the President's own office, so that its head could speak before Congress as well as report the findings and recommendations of all his own agencies to the President.

Dr. Brode has put his finger on a vital matter, of which Lord Hailsham has already shown himself to be fully cognizant. It is impossible to take too great care to ensure that what is involved in scientific policy is fully understood at all levels in the organization of government, both in the legislature and in the administration. While he is clearly not fully satisfied that, in the United States, a Department of Science would be an effective means of interpretation and formulation of policy, he firmly urges the establishment of a commission or study group to consider the organization of science and scientific policy in academic, industrial and government fields. Particularly this should examine the re-grouping of the Government's scientific agencies and activities, either under a Department of Science with a National Science Council, a National Science Institute or some other co-ordinated structure. Effective co-ordination would be required between such a re-grouping and those scientific agencies which remained separate, so as to ensure an efficient and comprehensive national science programme in the United States.

With this Dr. Brode suggests that there should be considered a re-alignment of the methods of distributing responsibility for support of fundamental research in educational institutions, possibly through the administration of university grants mainly by a department concerned with fundamental research, rather than by agencies concerned with application. He also suggests some separation of government-sponsored major research institutions from the educational and industrial system, especially of those institutions essentially concerned with applied science, and finally that scientists in government service should be linked with those in the academic field and in industry on a National Science Council. Primarily, he is concerned to see that creative thought, mature judgment and experience are brought to bear on these difficult problems of the formulation of scientific policy and the co-ordination of activities, and also on the attack on scientific and technical problems themselves.

Dr. Brode is undoubtedly right to maintain that thoughtful and penetrating analysis of the problem must precede any decision to change existing institutions and procedures ; and while he is prepared to support drastic action, it is only on the basis of full inquiry. That is known to be Lord Hailsham's attitude, and it is in this context that the papers presented last May at a symposium on Basic

Research in New York and now published by the American Association for the Advancement of Science* are of especial interest. Although fundamental research is only one element involved in the determination of scientific policy, it is probably the essential factor, and the one in which it is most difficult to determine priorities and to exercise co-ordination without detriment to scientific work.

The symposium, which was held at the Rockefeller Institute under the auspices of the U.S. National Academy of Sciences, the American Association and the Alfred P. Sloan Foundation, sought to examine facts concerning the support of basic research in the United States, the reasons why the nation did not give it adequate and suitable support, and ways in which the situation could be improved. Five specific questions were put to the invited participants. Is not the large support of applied research, and still more particularly the massive present support of development, in unhealthy relation to the meagre support for basic research ? Is it not true that industry pays eager lip-service to basic research, but in actual fact does not give adequate support to basic research, either within industry or elsewhere ? Has either industry or the Government learned how to protect basic research from the insistent demands of applied research and development ? Are not universities so deeply invaded by the demands for solving immediate problems, and by the temptation of income for so doing, that there are too few competent scholars pondering problems simply because it interests them to do so, so that scholars in the universities of the United States may have lost the manœuvring room for their continuing re-analysis of the universe ? Has it been effectively accepted that the spirit of basic research is an essential ingredient of the educational process—and that this fact should affect educational procedures at all levels ?

None of these questions can be dismissed as irrelevant in a British context, and although the answers which might be given may differ considerably, especially in detail, it is a tribute to the quality of the symposium that the addresses reproduced in this volume should bear so closely on current problems of the organization of the research effort in Britain which the Advisory Council on Scientific Policy must now be considering and which impinge closely on the expansion of universities and of technological education. That might indeed be expected from the eminence of the contributors, which include Dr. J. R. Oppenheimer, Dr. A. T. Waterman, Dr. L. A. DuBridge, Dr. J. R. Killian, Dr. C. H. Greenewalt, Dr. A. V. Astin, Dr. J. B. Fish and Dr. M. A. Tuve, and it is noteworthy that in directing the attention of the symposium at the start to the need for new knowledge, Dr. Oppenheimer was not content merely to urge that getting new knowledge was an ennobling as well as a useful activity. Beyond its character as a human, creative activity, he pointed to the wide field of study which man's institutions, his whole society now present—underlining, as it were, the basic contention in Dr. Brode's address already noted.

Dr. W. O. Baker, discussing the paradox of choice for the research worker, followed a very similar line suggesting that it is time we began to consider what specific skills as well as ideals are needed for the advancement of civilization. If a fraction of the effort that has been given to the acquisition of physical skills were given to the practice on proper levels of mathematical reasoning and exercise, we might prepare that small but necessary group which would interpret, as well as the essential group which would create, conceptual science to come. Dr. Baker quoted Dr. Warren Weaver's remark that "our society has not yet evolved a satisfactory way of fostering creative scholarship" in illustrating the potential contribution of scientific method in developing the appropriate institutions for both fundamental and applied research.

The clearest picture of the actual position of fundamental research in the United States was presented by Dr. A. T. Waterman, who, taking the figures for 1956–57, when some 7 per cent of the 9,000 million dollars expended in the United States on research and development went to basic research, said that of 300,000 full-time engineers and scientists estimated as engaged in research and development 27,000 were on fundamental research, 14,000 being in educational and other non-profit institutions, 9,000 in industry, and 4,000 in the Federal Government. Recognizing the vital importance of the universities in this field and the difficulties arising in regard to finance, Dr. Waterman concluded that the evidence shows that fundamental research requires stronger support at colleges and universities ; provision for renovation of research laboratories and for up-to-date scientific equipment presents outstanding problems. Clearly, the selection of problems and determination of priorities present very similar problems in the United States and in Britain, and Dr. Waterman seems more confident than Dr. Brode that the new organization in the United States is appropriate to the conditions there. He concedes, however, the importance of the right choice of men, and this is the factor that other participants repeatedly stressed. Dr. Killian remarked that a report recently presented to the new Federal Council for Science and Technology had cited highly trained scientific man-power as constituting the real limit to the development of materials research, and urged the need not simply for more fundamental research but also for more men capable of creative contributions. Dr. C. H. Greenewalt, who claimed that the du Pont de Nemours Company is now spending more than 15 million dollars a year on fundamental research, urged a substantial increase in unrestricted funds for the universities, provided both by industry and government, but based not upon projects but on men—not upon desired objectives but upon creative talent. Dr. M. A. Tuve, of the Carnegie Institution of Washington, commented that the United States has not had much success, as a nation, in encouraging quietly creative scholarship and intensely personal activity in research; and he suggested that the support of fundamental research is the support of ideas. This always means support of a creative investigator, and he proposed that public funds should be used to pay the salary for the remainder of his working life of a gifted research worker, who had clearly established by the age of thirty or thirty-five his creative ability.

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* Symposium on Basic Research. Edited by Dael Wolfle. (Sponsored by the National Academy of Sciences, the American Association for the Advancement of Science, and the Alfred P. Sloan Foundation, and presented at the Rockefeller Institute, May 14–16, 1959, New York City.) Pp. xx+308. (Washington, D.C.: American Association for the Advancement of Science ; London : Bailey Bros. and Swinfen, Ltd., 1959.) 27s.

Dr. Tuve visualized the creation in one decade of a body of some 500 or 600 outstanding investigators by the allocation of perhaps 40–60 million dollars a year in creating research professors or scholars of this type; and although in summarizing the symposium, Dr. D. Wolfle recognized that there are both administrative and financial difficulties, he pointed out that there are already precedents in the research professors at universities and the permanent staff of private research institutions. The participants of the symposium, in fact, recommended wider use of research professorships or similar posts as a means of furthering fundamental research.

The other recommendation which came from the symposium was that universities must always be expected to carry the largest responsibility for basic research, and that accordingly they should be provided with larger unrestricted funds. There was also repeated insistence that a larger proportion of the present Federal research and development budget should be available for the universities on an unrestricted basis. Only with a sufficiency of such funds can the universities adequately discharge their continuing responsibilities for teaching and creative scholarship, and maintain a reasonable balance between teaching and research and among the various fields of scholarship. It was equally plain that those present entertained the same distrust of project grants as universities in Britain have of sponsored research and earmarked grants. The vital factor is to free creative scholarship from whatever limits its efforts. It is essential to concentrate on intellectual quality, and while the importance of freedom to investigate and to publish was generally recognized, there was less disposition to concede that the difference between State-owned and privately owned institutions is always as great as is commonly maintained. Dr. DuBridge, in fact, was deeply sceptical of scientists as administrators, particularly in government institutions and when controlling the activities of other scientists through the administration of research grants.

This is not to challenge the freedom of the scientist to decide the conduct of his own research or the choice of his programme: it is rather to challenge the validity of attempting to apply cost-accounting practices too closely, and to stress the need to maintain the appropriate intellectual environment and the conditions of service under which basic research is most effectively pursued. That, as Dr. Fish pointed out, involves a certain freedom in the choice of problems, subject to the criterion of relevance, as well as freedom to carry a project to the point of demonstrating the merit of an idea and then to drop the study and follow the next good idea. These are not, however, inherent rights of individuals, and Dr. Tuve made it clear that the Carnegie Institution of Washington interprets the support of ideas always as the support of a man. Dr. DuBridge saw the essential problem as that of attracting sufficient of the nation's ablest minds to the universities and giving them full encouragement and support in pursuing whatever lines of inquiry interested them.

Admittedly that is a difficult challenge to meet, especially with the mounting costs of equipment and assistance in many fields of inquiry, and even though, in comparative costs, expenditure on academic fundamental research may not, in the United States, be more than twice what it was before the War. Thus the expenditure of fully 800 million dollars, which Mr. R. E. Wilson is inclined to regard as the current total expenditure on basic research in the United States, may be inadequate; and the determination of priorities and the establishment and maintenance of appropriate conditions are unlikely ever to be other than major pre-occupations in the United States as in Britain. The symposium threw up few, if any, novel ideas; but it demonstrated the way in which fundamental and applied research are interlocked and are also related to education and to professional training. The universities and the independent research institutions as well as industry and government institutions have their contribution to make, but in determining the structure in which all these activities are most appropriately fostered and co-ordinated the first consideration should be the promotion of excellence, the encouragement of able investigators and their sustained and adequate support with finance, equipment and assistance.

GALILEO AND EULER AS POPULAR EXPOSITORS

Discorsi e Dimostrazioni Matematiche Intorno a Due Nuove Scienze

Di Galileo Galilei. A Cura di Adriano Carugo e Ludovico Geymonat. (Classici della Scienza, 1.) Pp. xxv+887. (Torino: Paolo Boringhieri, 1958.) 7,000 lire.

Lettere a Una Principessa Tedesca

Di Leonhard Euler. A Cura di Gianfranco Cantelli. (Classici della Scienza, 2.) Pp. xxvi+960. (Torino: Paolo Boringhieri, 1958.) 8,000 lire.

THESE two volumes provide, among many other things, an excellent opportunity for comparing Galileo and Euler as popular expositors, albeit at greatly different levels. Galileo's "Discorsi" first appeared in 1638; published at Leyden, in order to evade the ban imposed by the Inquisition, they incorporate material on which the author had been meditating for a long time, and which is so varied that neither Galileo nor any of his friends succeeded in finding a suitable title for it. Thus the first of the four discussions into which the main portion of the book is divided actually begins with one of the principal themes—the strength of materials—but soon passes to an examination of the concept of continuity and thence to an essay on transfinite numbers.

The dialogue form in which the book is cast lends itself to, and indeed encourages, this sort of discursiveness. Galileo has distributed his exposition and commentary among three speakers: two of these, Salviati and Sagredo, are former pupils, while the third, Simplicio, is an imaginary personage. It is Salviati's function to enunciate and expound; Simplicio, on the other hand, represents the opposition; and Sagredo, an intelligent layman, not only provides the pretext for a further explanation, in simpler terms, of the author's ideas, but also is used to express concepts for which Galileo did not wish to accept full responsibility.

To obtain an adequate notion of the book's contents, one would have to quote Galileo's own index. The second main theme is the theory of accelerated motion, with application to projectiles; but intertwined with this is a considerable amount of applied mathematics as well as many pioneer results in the integral calculus.

The editors have carried out their task with great skill and devotion; the text, which runs to nearly 600 pages, is accompanied by 300 pages of notes. Moreover, those demonstrations which originally appeared in Latin are here reproduced with parallel translations in Italian. With regard to style, Galileo's own limpid prose furnishes a model which, unfortunately, few of his compatriots have seen fit to adopt.

Euler's "Letters to a German Princess" are a classic of a quite different kind; composed during 1760–62, they were addressed to his pupil, daughter of a Margrave, from whom he was separated by the vicissitudes of war. The letters were first published in French, at St. Petersburg, achieving an immediate and extraordinary success. Translations into most European languages soon followed, and numerous reprints (eight in Paris and six in New York) bore witness to the popularity which the work enjoyed for nearly a century, long after it was largely outmoded: suffice it to remark that the book contains an attack on Leibniz's theory of monads and a defence of Newton as against Descartes—both highly topical matters at the time when Euler wrote.

The reasons for this popularity are very clear. To begin with, the letters give a detailed description of almost the whole of elementary mathematics and physics, including astronomy, as Euler understood them. This part of the work is a masterpiece of exposition; it includes accounts of what were then recent results, such as the first experiments with electricity and the determination of longitude at sea (incidentally, for contributions to this problem Euler himself received a prize of £3,000 from the British Parliament). As a popular scientific encyclopædia the book would be difficult to surpass. But there is obviously another cause for Euler's success: the purely scientific information is coupled with instruction in formal logic, philosophy and religion, designed to combat the spirit of the Enlightenment. In this field Euler was an amateur, and an ill-informed one at that; he made himself ridiculous to his contemporaries.

A perplexing question raised by the "Letters" (234 in all) is the intellectual status of their recipient; one is forced to conclude that she was ignorant of almost everything—there are even lessons in elementary arithmetic—but deemed capable of absorbing anything whatever.

The present version, based on the second French edition, reads very well. Both the Galileo and the Euler are handsome books, and should be in every scientific library.

L. Roth

THE LUNAR SURFACE

Strange World of the Moon

An Enquiry into Lunar Physics. By V. A. Firsoff. Pp. x+226+12 plates. (London: Hutchinson and Co. (Publishers), Ltd., 1959.) 25*s*. net.

PERSISTENT observers of the Moon, many of them amateurs with small instruments and little scientific training, have often reported changes of various kinds in the appearance of the lunar surface; these have tended to be met with scepticism by most professional astronomers; but until the last year or two very few professionals have spent much time on lunar observations, and there is thus a considerable chance that genuine phenomena have been missed. The present book, which attempts to interpret changes seen by the author and other observers in terms of known physical and chemical laws, is timely in view of the indisputable observation by N. A. Kozyrev of a short-lived bright cloud of dust and of C_2 and C_3 gas near Alphonsus in November 1958, and in view of the increased interest in the Moon resulting from the new facilities offered by space rockets.

The author points out that the Moon's orbital plane is inclined at only 5° to the ecliptic, which, together with its large size, suggests that it is a captured planet rather than an off-shoot of the condensing proto-Earth. This leads him to speculate that, as a result of planetary perturbations and tidal action, the Moon may have approached and receded from the Earth several times, each approach having led to distinct mountain-building fault systems in both bodies; but an elementary mathematical treatment given in an appendix shows that rather extreme conditions must be assumed. He rejects the impact theory for the origin of craters in favour of 'lunavoes' (a term coined by J. E. Spurr), which are essentially broken bubbles of gas bursting out from a contracting surface; wrinkle ridges on certain maria are treated as evidence for a viscous liquid filler, rather than dust. Mr. Firsoff attributes ray systems to erosion by running water, fed from large deposits of ice accumulated below the spongy surface rocks, and he suggests that the maria, also, are filled by impure ice. The main changes observed consist of the occasional disappearance and creation of craters, puffs of dark and bright dust or vapour, and local changes in brightness and colour at sunrise, many examples of which have been studied by Firsoff himself with the aid of filters, and which he attributes to the evaporation of snow (or possibly carbon dioxide and other volatile solids). This leads on to a discussion of the lunar climate and atmosphere in which it is shown that, in view of Lyman Spitzer's work on the terrestrial atmosphere and of the presence of appreciable amounts of argon resulting from the decay of potassium, the old idea that all water vapour would have escaped may be quite untrue. Firsoff concludes that the lunar surface, or at least subsurface regions, can be the seat of extensive climatic and seasonal changes, and even of rudimentary forms of life.

The ideas presented in the book are interesting and highly provocative, and the significance of much of the experimental and theoretical work that has been done is discussed in considerable detail, with helpful technical details in the appendixes. Basically, however, the author's arguments only show that his ideas are barely possible; he has certainly not shown that they are at all likely. In particular, the idea that water is present on the lunar surface is highly implausible, not because of escape, but because of the presence of solar ultra-violet radiation, which is known to dissociate water vapour into hydrogen and oxygen in the upper atmosphere of the Earth; the resulting hydrogen would certainly escape from the Moon. Again, the discussion of craters and mountains fails to do justice to the very strong and detailed case for meteorite impact that has been made by R. B. Baldwin and T. Gold. The value of the book thus lies more in its description of observations and in the questions it provokes than in the answers that it suggests. B. E. J. Pagel

SECONDARY MODERN SCHOOLS

Your Secondary Modern Schools

An Account of Their Work in the Late 1950's. By J. Vincent Chapman. Pp. 298. (London: The College of Preceptors, 1959.) 12*s*. 6*d*. net.

Although many opinions are given about the encouraging successes or the utter inadequacies of the secondary modern school, these are often coloured by information gathered from one or two areas only. As a result many educationists, including heads and assistants from secondary modern schools themselves, have a distorted picture of the general position in the 3,500 secondary modern schools scattered throughout Great Britain.

Benefiting by the award of the Doreck-Isbister Fellowship by the College of Preceptors, J. V. Chapman has been in touch with about half the total number of schools and here records the information and impressions he has gained from his face-to-face and written inquiries. Chapman sought information under three headings. First, on the amount of time allowed for a certain subject on the school's timetable; secondly, the general syllabus of work; and thirdly, the titles of books being used in the main subjects. From this he has built up a record which, on the whole, is one of good work being done in discouraging circumstances, but which, given more buildings and adequate staffing, could be considerably improved.

The demands on school headmasters and headmistresses are heavy but cheerfully accepted. Her Majesty's inspectors have to be accommodated; parent-teachers' associations have to be stimulated; medical inspections have to be arranged; reports for parents and employers have to be written; school journeys need organizing. These and many extramural activities throw considerable strain on the school staff; but, even with school milk and school meals, the masters and mistresses carry on with the main job of providing an all-round general education for pupils of widely varying intellectual levels.

Despite the explosive outbursts of the 'Now-when-I-was-a-boy' school, English is well taught in the 3½ hr. a week which is allotted to it in the average school. The influence of good teaching, however, is soon minimized in homes which culturally know little more than the 'Daily Distress' and Channel 9. About the same time is allowed for mathematics; but here the quality of work done depends quite simply on whether or not the school has managed to capture and retain a competent teacher.

Science teaching is, by-and-large, non-existent. Apart from the odd school with a proud record, the story is one of laboratories waiting for teachers or of teachers, despairing of facilities which take a long time to come, leaving to go elsewhere. It is a sad reflexion that the author of such a book has to fill up a chapter heading with 'Science, including Gardening'. Subjects like history, geography and crafts are well covered although many more schools would teach metalwork if they had the facilities. Physical education is a credit to the schools; a beginning has been made with French and commercial subjects. Domestic science and needlework are given considerable attention in girls' schools, while, in some schools, music is treated with such enthusiasm that "the bugbear of the breaking voice is overcome simply by denying that it exists".

The findings in this factual account of what is going on in secondary modern schools to-day inevitably leads one to question the wisdom of the Crowther Committee in recommending that the school leaving age be raised compulsorily for all children to sixteen. Before this is done the nation should repay the considerable debt to the worthy men and women in secondary modern schools who have managed to make such good bricks with so little straw. Any money available for education should be spent on better buildings, providing adequate staff, and seeing that no longer should some headmasters be able to write that in each year they are allowed to spend a total of 9*s*. 6*d*. on books for each child. Two packets of cigarettes a year! T. H. Hawkins

"B.D.S." PHYSIOLOGY AND BIOCHEMISTRY

Textbook of Physiology and Biochemistry

By Prof. George H. Bell, Prof. J. Norman Davidson and Prof. Harold Scarborough. Fourth edition. Pp. xi+1065. (Edinburgh and London: E. and S. Livingstone, Ltd., 1959.) 63*s*. net.

This text-book, like an old friend, is often referred to by initials rather than by name, and to many students, even dental students, "B.D.S." probably first suggests this book rather than a degree. This new edition comes only three years after the last one—an indication of the popularity of the book—and it is good to see that Spanish and Italian editions are now being published. It would scarcely be expected that many major changes would have been made after so short a time, and the chapter headings are the same as before, except that a chapter on glycolysis and the citric acid cycle replaces one which summarized liver function. Many changes, though, have been made within the chapters to bring the text and bibliographies up to date. The chemical structures are, in many cases, more clearly set out, and there are some new illustrations, including an attractive coloured one of protein chains. This book has always been notable for its generous provision of illustrations, and students appreciate the way in which so much of the experimental material is presented graphically so that it can be more quickly grasped and more easily remembered. The experimental evidence for the facts is given so far as possible, and this is one of the most valuable qualities of the book.

I was disappointed not to find any discussion of the interesting genetic and biochemical features of abnormal hæmoglobins such as the one associated with sickle cell anæmia, nor anything about the trial of diets low in phenylalanine as a possible means of preventing brain damage in infants suffering from phenylketonuria. In a book used so much by medical students some of the over-generous space given up to structural formulæ of, say, the porphyrins might be better used in some such way. This is not a plea for a larger volume: indeed, the authors are to be congratulated on having made this edition slightly smaller than the one before—something that rarely happens in a new edition. It is greatly to be hoped that they will maintain their enthusiasm for keeping this excellent text-book up to date. If at the same time they find it possible to continue the pruning process, so much the better; future generations of medical, dental and science students will be the more grateful to them.

D. C. Harrison

METEOROLOGY FOR GEOGRAPHERS

Weather and Climate

By Dr. Clarence E. Koeppe and Dr. George C. de Long. Pp. viii+341. (London: McGraw-Hill Publishing Company, Ltd., 1958.) 58s.

THE authors of this book are American professors of geography. They state in the preface that it is intended primarily for students of geography needing a more detailed and scientific knowledge of weather and climate than is provided in a first geography course, but lacking the knowledge of physics and mathematics required for a standard course in technical meteorology, and for students interested in a liberal arts training without the intention of becoming professional geographers or meteorologists.

The first eleven chapters set out to give in 144 pages an outline of physical and dynamical meteorology without using any mathematics or assuming any knowledge of physics. This is a very difficult task in which the authors have not succeeded. The sequence of ideas is loose, the essential physical principles are not stated or not clearly brought out, and in places the text would certainly be misleading or puzzling to readers of the type to whom it is directed. What, for example, is to be made of the following sentence about the relation of winds to the contours on the 500 mb. chart, "Winds tend to blow parallel with the contours and thus toward the depressions, which is in accord with the principle, already stated several times, that although air moves down the isobaric slope, it can move at a deflection of as much as 90 degrees to the slope"?

The second part, of 190 pages, on climate is much better though not entirely free from error. The Earth's climates and corresponding types of vegetation are described in fair detail in terms of the Koppen-Geiger classification of them. An attractive feature is the photographs of typical landscapes in the various climatic types. Most illustrations, including cloud forms, are well reproduced, but what would the late G. A. Clarke of Aberdeen, Scotland, have said to the 'acknowledgments' (to the U.S. Weather Bureau) for the two of his photographs in Figs. 11–2 and 11–3 or the quality of reproduction of the first?

The book cannot on the whole be recommended.

G. A. Bull

ANIMAL MYCOSES

Fungal Diseases of Animals

By Dr. G. C. Ainsworth and P. K. C. Austwick. (Review Series No. 6 of the Commonwealth Bureau of Animal Health.) Pp. xii+148+12 plates. (Farnham Royal: Commonwealth Agricultural Bureaux, 1959.) 20s.

CLINICAL mycology has never been a favourite subject among pathologists. Lecturers promise to deal with it at the end of the course, time permitting; the hospital bacteriologist unwillingly allows a few mould cultures to lie at the end of the bench until one day, angered by the inexplicable appearance of a green flannel-jacket of *Penicillium* on some precious batch of media, he rises up and indignantly consigns them to the autoclave. Identifying and classifying moulds is a formidable enough task to account for much of the unpopularity of the subject; another cogent factor is the shortage of reference works. Conant's "Manual of Clinical Mycology", published in 1954, relieved the situation as regards fungal diseases of man, and Ainsworth and Austwick have now attempted to codify current knowledge of veterinary mycology. The literature is extremely well covered, and for this reason the book should be of great value to research workers and teachers; undergraduate students (for whom it is said to be partly intended) will probably find much of the detail confusing, and if too little space is devoted to clinical matters for the book to have any great appeal for the practitioner, this is perhaps understandable since neither author is a veterinarian.

It is the contrast between the general ætiological patterns of animal and human mycoses that medical workers and biologists will find most fascinating. The key to the problem seems to be the greater exposure of domestic animals to unhygienic conditions, dirt, dust and spoiled or partly decomposing foodstuffs—in short, to infection with normally saprophytic fungi. Thus aspergillosis of chicks may follow inhalation of dust from mouldy grains or litter; bovine mycotic abortion is associated with the winter feeding of spore-containing hay, mycotic mastitis with the feeding of stale brewers' grains and mould toxicoses with damaged maize and other cereals; in sheep the multiplication of saprophytic Actinomycetes in rain-soaked and rotting wool leads to mycotic dermatitis, and soil or mud contamination of the limbs is a major factor in the ætiology of cattle farcy, a form of nocardiosis recalling in many ways Madura foot in the human subject. This is a theme which might well be elaborated in a later book, and who better than Dr. Ainsworth with his unique knowledge of both medical and veterinary mycology to do so?

J. E. Smith

Beetles of the British Isles

By E. F. Linssen. First Series: The Superfamilies Caraboidea, Palpicornia, Staphylinoidea and Diversicornia. Pp. 300+39 plates. Second Series: The Superfamilies Clavicornia, Heteromera, Lamellicornia, Phytophaga and Rhynchophora, and including the Strepsiptera. Pp. 295+87 plates. (Wayside and Woodland Series). (London and New York: Frederick Warne and Co. Ltd., 1959.) 60*s*. net the set.

THE aim of the "Wayside and Woodland" series is to enable naturalists and others to identify animals and plants in the field, chiefly by comparing their specimens with good-quality figures. In the present work the figures of adults are taken mainly from Fowler (1887–1891) and those of larvæ (as usual) from Schiodte (1861–1883). The latter have been adapted by Mr. A. F. Stuart, who is also responsible for the text-figures and the colouring of the plates. His work, original or adapted, is excellent. Of the 126 plates, 64 are coloured.

Many beetles can only be examined satisfactorily under a microscope, and in this work fine structural characters are given so that closer and more reliable determinations can be made than by using figures alone. There are keys to the families of some major groups. The system of classification adopted is that of Kloet and Hincks (1945). A most useful table is given in both volumes showing the superfamilies of Polyphaga recognized by various authors (1 : 140–141 and 2 : 16–17).

A work of this kind must inevitably take the form of a catalogue of species, with notes, and the author should be able to express himself clearly, concisely and authoritatively. Unfortunately, Mr. Linssen's style is cumbrous and many of his statements are inaccurate or obscure. Particularly deplorable is the large number of misspellings of scientific names which cannot fail to confuse the amateur, especially as genuine obsolete spellings are often given.

R. T. THOMPSON

From Ape to Angel

An Informal History of Social Anthropology. By H. R. Hays. Pp. 461+32 plates. (London: Methuen and Co., Ltd., 1959.) 36*s*. net.

BOOKS to popularize science may take many forms. Very attractive are those where some great man of science takes a recent discovery and translates it into language which can be understood by those whose knowledge of science is limited, but who still have inquiring minds. Equally valuable is the general interpreter, a man who appreciates the interest of the general public in a particular aspect of science at a given time, and distils knowledge from a wide variety of sources and presents it in an attractive and readily assimilable form. Because these works contain little that is wholly new and, for the sake of simpleness, sometimes put considerable strain on veracity, they are often dismissed with contemptuous references to 'pot-boiling'. Hays's popular narrative of man as a social animal, including accounts of his primitive behaviour, could be put in the latter category. Yet here, to match up with the considerable interest in the lives of primitive peoples which has been evoked by television, is a book which may well develop into a best-seller. To describe the extraordinary ethical, religious and marital customs of primitive peoples, Hays provides a short biography of the lives of famous pioneer anthropologists such as Henry Shoolcraft, Westermarck, Sir James Frazer, Franz Boas, Bronislaw Malinowski and others. The result has been the production of an informal history of social anthropology which is attractive and informative and which does the author credit for the assiduous way he has collected his information and put it together. "From Ape to Angel" has deservedly won recommendation from the Book Society.

T. H. HAWKINS

The Thyroid Hormones

By Dr. Rosalind Pitt-Rivers and Jamshed R. Tata. With a Chapter on Diseases of the Thyroid by W. R. Trotter. (International Series of Monographs on Pure and Applied Biology. Division: Biochemistry, Vol. 1.) Pp. xiii+247. (London and New York: Pergamon Press, 1959.) 50*s*. net.

A BOOK by two such authorities on research into the thyroid gland could scarcely fail to be useful. The authors say that their aim is to unfold the story of the thyroid hormones and to discover which hypotheses concerning their biosynthesis, mode of action, and the ætiology of thyroid disorders, have received experimental support. They also have tried to provide research workers in the field of thyroid physiology and biochemistry with as many references to the recent literature as possible.

The unexpected appearance of tri-iodothyronine on the scene in 1952, in which Dr. Pitt-Rivers herself played a leading part, has led to much new thinking about thyroid hormones. We are still without a finally accepted view about the nature of thyroid hormones and their mechanism of action, but the present book sets out as much as can be reasonably concluded at the present time. More information about the actual amounts of the different thyroid hormones found in blood and tissues under physiological conditions might have been expected, but undoubtedly much checking and re-checking must be done before final conclusions are drawn. The authors have clearly succeeded in their formulated aims in writing this book.

F. G. YOUNG

Dana's Manual of Mineralogy

Revised by Prof. Cornelius S. Hurlbut, jun. Seventeenth edition. Pp. xii+609. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1959.) 78*s*. net.

JAMES DWIGHT DANA'S classic "System of Mineralogy" has stood the test of time, so also has his "Manual of Mineralogy", which has now reached its seventeenth edition. The first edition of the "Manual" appeared in 1848; since then many revisions and editions have appeared, the last being in 1952. The latest revision was undertaken by Cornelius S. Hurlbut, jun., professor of mineralogy in Harvard University. It introduces the broad use of the crystal-chemical approach, and many of the chapters have been expanded in this light. The chapter on "Crystal Systems" now includes discussions of the stereographic projection, the thirty-two crystal classes, calculation of axial ratios and X-ray crystallography. This new approach enables the properties of minerals to be treated through fundamental structures and chemical consideration. It is hoped that if future revisions are made care will be taken not to resolve the "Manual" into a form indistinguishable from the "System of Mineralogy". It is better that the "Manual" maintains its original purpose, for students, amateur collectors and mining geologists.

INCREASES IN THE LOCAL GAMMA-RAY BACKGROUND DUE TO NUCLEAR BOMB FALL-OUT

By J. VENNART

Radiological Protection Service, Belmont, Sutton, Surrey

IN 1956, prior to the large-scale testing of thermonuclear weapons, Spiers and Griffith[1] and I[2] made measurements of the local gamma-ray background in Leeds and Aberdeen, and in Belmont and London respectively. A noteworthy feature of the measurements was that, due to the low radioactivity of the underlying chalk of the North Downs, the local gamma-ray background to which man is exposed in Belmont was about 60 per cent of that in Leeds. It is now known that this is principally due to the small amount of potassium in chalk (about 20 parts per million compared with 5 per cent in common building materials), giving a correspondingly low concentration of the naturally occurring radioisotope potassium-40. In addition, the contribution from the other principal naturally occurring radioelements, thorium and uranium, is about ten times less than that in common building materials[3].

These facts have had important repercussions with regard to the design of low-background laboratories within the chalk at Belmont, which are used for the measurement of low levels of radioactivity in humans[4] and in food[5]. We are therefore concerned about any increase in the local gamma-ray background of our environment, and accordingly in April 1958, shortly after our low-background laboratory was completed, I initiated regular checks out of doors at the Belmont Hospital and have continued these to the present day. In addition to these measurements I have also on one or two occasions repeated the measurements reported for 1956 inside and outside a dwelling house at Belmont and at other spots in and around the Hospital. The results show that during the period of the measurements, the local gamma-ray background out of doors increased steadily and reached, in April 1959, a maximum which was about twice that in 1956. In recent months there has been a spectacular fall, presumably due to the cessation of nuclear testing, and at the present time the local gamma-ray background out of doors is barely significantly different from that measured in 1956. As regards the local gamma-ray background inside buildings, measurements made on two occasions indicated that the increase varied considerably with the shielding effect of walls but might be in the order 50 per cent of that reported out of doors. I am reporting these measurements now so that a comparison may be made with others reported recently by Peirson and Salmon[6] for various parts of Britain and by Spiers[7] for Leeds, and also so that the magnitude of this increase in relation to man's previous radiation history may be discussed.

The measurements were made, as they were in the early experiments[2], with two counters G.10 lead and G.10 carbon enclosed in brass cases 1½ in. in diameter and 1·2 mm. thick. The same G.10 carbon counter was used for the whole period, including the 1956 measurements, whereas, due to breakages, it was necessary to use four different G.10 lead counters. However, these had previously been calibrated against a radium standard and their sensitivities were the same within 2 per cent. All the counters were checked regularly for constancy of response with a weak radium source. The residual counter response to cosmic radiation and counter contamination was determined by shielding experiments out of doors and also within the 9-in. thick steel enclosure used by Spiers at Leeds. The value obtained was approximately 40 counts/min. for either type of counter. As described in the earlier paper, the ratio of the counting rates due to local gamma-rays from the two counters at a given site is a measure of the relative proportions of primary radiation and scattered radiation in the local gamma-ray background. This ratio has remained substantially constant at about 2·1 for all sites throughout the whole series of measurements. It may then be shown[2] that the local gamma-ray dose-rate in m.rads/yr. is given very approximately by (1) counting rate due to local gamma-rays in the G.10 lead counter multiplied by 0·9, (2) counting rate due to local gamma-rays in the G.10 carbon counter multiplied by 1·9.

Fig. 1 shows the net counting rate out of doors due to local gamma-rays (left-hand ordinate), for the G.10 lead counter placed about 3 in. above closely mown grass outside the low-background laboratory. Apart from December 1959, when only two measurements were taken, about forty individual measurements were made each month; for any single measurement the standard deviation of the counting was about 2·5 counts/min. The mean value for any day was found from three to four separate measurements of the background; and the graph shows, for each month, the mean and the standard deviation of the distribution of these daily mean values. An interesting feature of the measurements was the increase in counting rate which followed heavy rain. For example, in August 1958, during a heavy thunderstorm, the counting rate due to local gamma-rays rose to 85 counts/min., but within 4 hr. of the rain ceasing this had fallen to about 42 counts/min. A large fraction of the difference, 43 counts/min., may therefore be attributed to the gamma-emitting decay products of the naturally occurring gas radon which decay with an effective half-period of the order of half an hour. No measurements which were taken during or immediately following heavy rain have been included in the graph.

The graph also shows (right-hand ordinate) the dose-rate in m.rads/yr. using the calibration factor given above. Although these values represent the dose-rate at 3 in. from the ground, other measurements made at a height of 2 ft. gave similar values. This result is in accord with the well-known fact that the local gamma-ray dose-rate is independent of height above the surface of the Earth, provided this does not exceed a few metres[8]. Accordingly, the dose-rate values shown on the graph are, to a close approximation, the local gamma-ray dose-rates to which man has been exposed at the site.

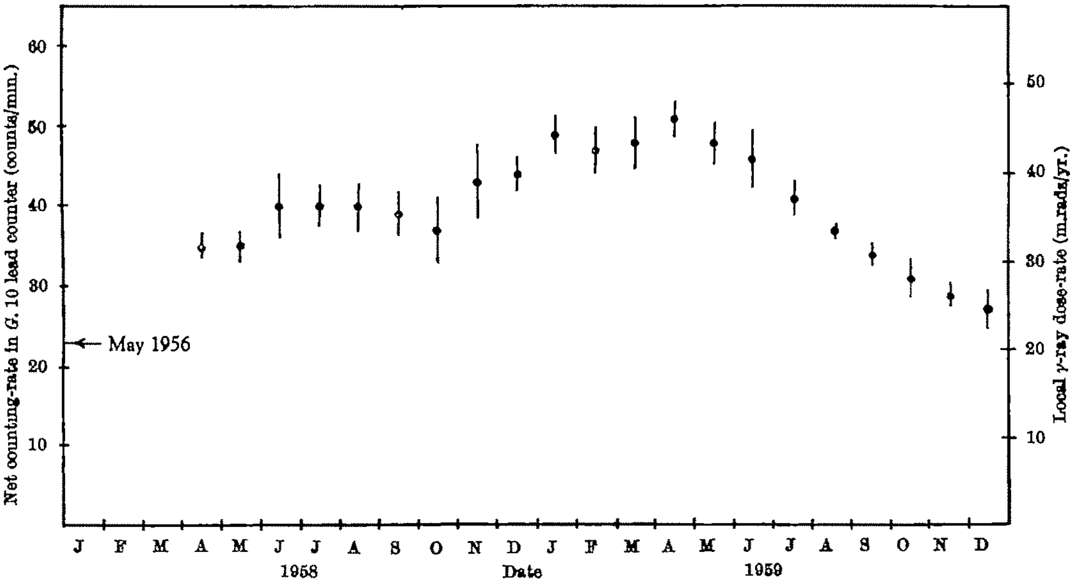

Fig. 1. Local gamma-ray background out-of-doors at Belmont, Surrey

Measurements were also made with the carbon counter on five occasions throughout the series, and the dose-rate so determined agreed with that obtained from the *G*.10 lead counter to better than 5 per cent.

It will be seen from the graph that in April 1958, when the present series of measurements began, the counting rate due to local gamma-rays was 12 counts/min. greater than the value, 23 counts/min., obtained in May 1956 at the same site. Measurements at two other sites, one over a tarmac courtyard within the Belmont Hospital grounds and one over the garden of a house in Belmont, at which measurements had been made in early 1956, showed similar increases in counting rate, namely 15 and 11 counts/min. respectively. It is of interest to note that the increase in counting rate was about the same over closely mown grass, cultivated garden and tarmac. This evidence, coupled with that of Spiers[7], would imply that it may be over-optimistic to use a protection factor of 3, as was suggested in Appendix *M* to the Medical Research Council's report of 1956 [9], to take account of fall-out material which is washed into drains or otherwise removed from the topmost layer of the surface of the Earth. From April 1958 the counting rate continued to increase steadily, apart from a fall in October 1958, to a maximum in April 1959 which was more than twice the 1956 value. The corresponding dose-rate at this peak period was about 25 m.rads/yr. more than in 1956. By December 1959 the counting rate due to local gamma-rays was barely significantly different from that in 1956. The dose-rate outside the house in Belmont was also barely significantly higher than in 1956. The effect of the series of tests during 1957–58 and especially in October 1958, as well as the cessation of all tests since that time, is clearly demonstrated.

From spectrometric measurements we have made of the radioactivity in the air during the period, there can be little doubt that the increased counting rate was due principally to gamma-emitting fission products of relatively short half-lives, less than one year, namely zirconium-95 and its daughter niobium-95, together with ruthenium-103 and rhodium-106. The longer lived cæsium-137 would be present to a very much smaller extent due to the much lower fission yield. The variation of counting rate with time shown on the graph is in fact very closely in step with that shown by Peirson and Salmon[8] for the cumulative deposition of fission products at Milford Haven. The maximum increase in the local gamma-ray dose-rate, 25 m.rads/yr., which occurred during April 1959, is the same as that obtained by Spiers during May 1959 over a lawn in Leeds[7]. It is also very close to the value which may be calculated for Belmont, where the mean annual rainfall is about 25 in., on the observed relationship between rainfall and cumulative fallout[8].

Using both counters, the local gamma-ray background was measured indoors during April 1958 in one ground-floor laboratory at the Hospital and at several sites on the ground-floor of the house in Belmont. There was no significant increase above the values obtained in 1956 for the room in the Hospital and the average increase in the house amounted to about 4 m.rads/yr., which was about equal to the standard deviation of counting the difference. The maximum increase in the house was about 8 m.rads/yr., compared with about 10 m.rads/yr. out of doors at the same time. Large variations in the local gamma dose-rate indoors are of course expected due to the different shielding effects of walls and windows. For example, in the Hospital, where no increase in dose-rate was measured, the counter was protected from fission products on the ground outside by brick walls 20 in. thick. It is clear, however, that it might be unduly optimistic to assume a protection factor of 7 in all cases for the shielding effect of buildings, as was suggested in Appendix *M* to the Medical Research Council report[9]. It was unfortunately not convenient to make measurements indoors during April 1959, the peak period for the outdoor measurements, but the presence of fission products indoors at this time was clearly demonstrated by measurements of the gamma-ray spectrum from the refuse from a vacuum cleaner which was being used at the house in Belmont. By November 1959, the local gamma-ray dose-rate in the house was not significantly different from that measured in 1956, demonstrating that any fission products inside the building had decayed to a negligibly low level, in step with the conditions out of doors.

The significance of an increase in the local gamma-ray dose-rate outside of about 25 m.rads/yr. has been discussed very fully by Spiers[7]. It will be clear that the effective increase in man's radiation status is markedly dependent on the assumptions which are made with regard to the protection factor afforded by wash-off into drains and into the deeper layers of the soil, by the shielding effect of buildings and by the fraction of the day spent out of doors. The measurements reported here would indicate that the protection factors assumed in Appendix *M* to the Medical Research Council's report of 1956[9] with regard to the first two are possibly over-optimistic. There may also be large numbers of people for whom a period of 2½ hr. out of doors, the value used in the report, may also be optimistically low. In fact the measurements reported here are not inconsistent with the most pessimistic factors suggested by Spiers[7]; namely, that the dose-rate indoors due to fission products might be as much as 50 per cent of that outside, and that the dose-rate outside is roughly independent of the nature of the ground surface. If only 2½ hr. are spent outside each day, it would then follow that, by April 1959, the local gamma-ray dose-rate to which man was exposed, due to fission products in the environment, was about 14 m.rads/yr. greater than in 1956. Assuming that body tissues reduce the gonadal dose to two-thirds of this value[7], the genetically significant increase in the annual dose-rate would be about 9 m.rads. This is about 9 per cent of the total from the natural background radiation, which includes local gamma-radiation, cosmic radiation and radium and potassium-40 in the body[9]. If, therefore, testing recommenced and fallout continued at the same rate as 1957–58, the increase in man's gonadal dose from fission products on the ground might itself be greater than the total from all other man-made sources of radiation with the exception of that received from diagnostic X-rays[9]. It might be worth noting that plants and animals which are not protected by the shielding effect of buildings would receive additional gamma-ray doses about three times greater. They might also receive additional genetic doses from the beta-rays emitted by fission products, whereas the human gonads are protected from this source of radiation by the shielding effect of overlying tissues and clothing. The local gamma-ray background is, however, at the moment barely significantly higher than before large-scale nuclear testing began and, if there are no further tests, we might conclude that the increment of dose over the past three years is of small importance compared with, for example, the difference between the doses of natural background radiation experienced by persons in Leeds[1] and Belmont[8].

There will also be small additional doses to the whole body and the skeleton from internally deposited fission products, principally cæsium-137 and strontium-90 respectively. A report of our measurements of cæsium-137 in humans during 1958 and 1959 will be published shortly.

It is a great pleasure for me to record my appreciation of the work done by Miss S. Terry, who has been responsible for almost all the experimental work; amounting to about 1,000 separate measurements of the local gamma-ray background.

[1] Spiers, F. W., and Griffith, H. D., *Brit. J. Radiol.*, **29**, 175 (1956).
[2] Vennart, J., *Brit. J. Radiol.*, **30**, 55 (1957).
[3] Turner, R. C., *et al.*, *Nature*, **181**, 518 (1958).
[4] Maycock, G., and Vennart, J., *Nature*, **182**, 1545 (1958).
[5] Mayneord, W. V., *et al.*, *Nature*, **182**, 1473 (1958).
[6] Peirson, D. H., and Salmon, L., *Nature*, **184**, 1678 (1959).
[7] Spiers, F. W., *Nature*, **184**, 1680 (1959).
[8] Hultqvist, B., "Studies on Naturally Occurring Ionizing Radiations" (1956). Report of the United Nations Scientific Committee on the Effects of Atomic Radiation, 101 (1958).
[9] "The Hazards to Man of Nuclear and Allied Radiations" (H.M.S.O., 1956).

ANTIBODY PRODUCTION

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THE mechanism of acquired immunity is as much a mystery to-day as it was sixty years ago. The facts which have come to light since then have provided little basis for choosing between the ever-increasing number of theories of antibody production. Even aspects of Ehrlich's side-chain theory[1,2], which was dropped almost completely for a time as a result of Landsteiner's work, are now being discussed seriously again, especially in view of some recent suggestions on the nature of immunological specificity[3,4].

The starting point for most speculation and experimental work on the mechanism of acquired immunity is, not surprisingly, the production of specific circulating antibodies in mammals. However, the failure of this approach to bring us much nearer to understanding the processes involved suggests that perhaps more consideration should be given to the possible origins of the phenomenon of antibody production in evolution. Some speculation from such a phylogenetic point of view has resulted in the suggestions which are discussed in this article. Although the hypothesis which is formulated does not seem to have been put forward in its entirety before, a large part of it is constructed of elements of the previous theories and ideas of others[1–9].

One of the most impressive features of the immune mechanism in vertebrates is its capacity to distinguish between constituents of the body and components of other organisms. Since the ability to discriminate between 'self' and 'not-self' is a necessary attribute of the phagocytic cells of the simplest multicellular animals, it is reasonable to suspect that this particular characteristic of immunity goes back very far in evolution—to periods long before the phenomenon of antibody production came into existence. In other words, the capacity for self-recognition is the most elemental of all the characteristics of the immune response, and it is therefore all the more remarkable that some of the important antibody theories scarcely consider this aspect of the problem, while in others the explanation for it represents the most far-fetched and the least satisfactory part of the whole argument.

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It is becoming increasingly evident that the specific structure of molecules on cell surfaces in both unicellular and multicellular animals is an extremely important factor in connexion with the normal functioning and interaction of cells. Specific surface affinities or compatibilities are apparently responsible for a wide variety of cytological phenomena which involve adherence of one cell to another. For example, 'fertilizin', which coats the ova of various marine animals, has a specific affinity for the 'antifertilizin' on the surface of the sperm[10], and the re-aggregation of sponge cells which have been scattered mechanically is apparently due to the interaction of specific surface components[11]. It is also highly probable that surface 'antigens', characteristic for each type of cell, play a vital part in the organization of tissue in morphogenesis[12–14].

An interesting example of the importance of specific surface components in Protozoa, particularly in relation to feeding mechanisms, is provided by the case of *Entamoeba hartmonella* in which bacterial 'agglutinins' have been described[15]. If a suspension of this amoeba is mixed with certain flagellated bacteria, the latter can soon be seen 'clumped' on the surfaces of the Protozoa. Afterwards phagocytosis of the bacteria occurs. Apparently the fixation is due to the presence on the surface of the amoeba of molecules which have an affinity for components of the flagella of the bacteria.

It is not unlikely that in simple multicellular organisms in which phagocytes play such an essential part both in nutrition and in defence, similar surface factors may form the basis of the mechanism by which these cells recognize the foreign matter. We can envisage that the phagocytes are, like *Entamoeba hartmonella*, coated with molecules with structures complementary to bacterial components. However, for phagocytes to be effective in the defence of the animal against infection they must have an affinity for a much wider range of micro-organisms than does *Entamoeba hartmonella*. It is therefore necessary to assume the presence on their surfaces of a large number of different molecules of varying steric configurations.

Concerning the synthesis of these molecules, it is possible that the structural variation essential for the wide range of specificities might be achieved by the presence in the cells of a corresponding number of distinct ribonucleic acid templates. An alternative and somewhat more economical possibility is that only a single type of ribonucleic acid template is needed; but that the structure of the protein molecule is such that it is extremely versatile with respect to the number of possible shapes that it can take up as it folds after coming off the template (cf. ref. 6). The final specific affinity of each molecule would thus depend upon the shape taken up during this random folding process.

To account for the ability of the phagocytes to distinguish between 'self' and 'not-self', two possibilities can be considered. First, we can suppose that two types of cell are implicated in the process. One of these cells produces, and liberates into the body fluids, molecules possessing random affinities but all of which have also a second affinity for the surface of the phagocytes. Molecules in which the randomly determined specificity is complementary to self-components are soon 'neutralized' and removed from the body fluids. The others continue to circulate in the fluid until adsorbed by the phagocytes by virtue of their second and constant affinity for the phagocyte surface. Thus the phagocytes become coated with molecules complementary for many different 'determinant groups' other than those occurring on self-components. This idea of molecules with such 'double specificities' is not entirely fanciful, since it is known that sera of immunized mammals sometimes contain antibodies which have an affinity not only for the immunizing antigen but also for the host's own cells. Normal cells which have been treated with sera containing such antibodies are afterwards capable of specifically adsorbing the antigen[16–18].

An alternative possibility would be that the phagocyte, like *Entamoeba hartmonella*, synthesizes its own 'receptor' molecules, but that the production of these ceases before the cell reaches maturity and before it is able to 'phagocytose'. Molecules with an affinity for 'self' would become 'neutralized' early on, so that by the time the phagocyte reaches maturity, only 'not-self' affinities would remain.

To account for so-called 'natural antibodies' in invertebrates we can postulate the production and release into the body fluids of molecules with random specificities but with no special affinity for phagocytes. Molecules with reactive groups for self-components are quickly absorbed out, thus never reaching harmful concentrations. Only molecules with 'not-self' affinities would tend to accumulate in the fluids.

It is suggested, then, that protein molecules of random affinities, especially coating phagocytes, play an essential part in the processes of feeding and microbial defence in the primitive Metazoa. We can now turn to discuss the specific immune response of higher animals, taking the view that its mechanisms are basically similar to, and in fact evolved from, those which are responsible for the ability of invertebrate phagocytic cells to recognize and respond to foreign matter.

The first step in antibody production must almost certainly consist of the uptake of the antigen by some kind of cell, and although absolutely nothing is known of how this comes about, it is not difficult to imagine that the specialized antibody-forming cells are coated, as was postulated for the phagocytes of invertebrates, with 'receptors' for a wide range of foreign molecules (antigens). We may suppose that these receptors are closely related to, if not identical with, γ-globulins. The actual shape and hence specific affinity of each receptor molecule might again be determined solely by the random folding process which follows its release from the ribonucleic acid template.

The usual inability of individuals to respond immunologically to their own proteins can be explained simply by assuming that the antibody-forming cells possess no receptors for self-components. The absence of 'self-receptors' on the cells can be accounted for, as in the case of the invertebrate phagocytes, by supposing either that the 'antigen receptors' are produced elsewhere and so 'neutralized' by the body's own components before they reach the antibody-forming cell, or that they are produced by the immature antibody-forming cell itself, and 'neutralized' before the maturation and development of the full potentialities of the cell. In either case the antibody-forming cells will possess no receptors capable of reacting with any proteins present in the circulation before the antibody-forming system itself

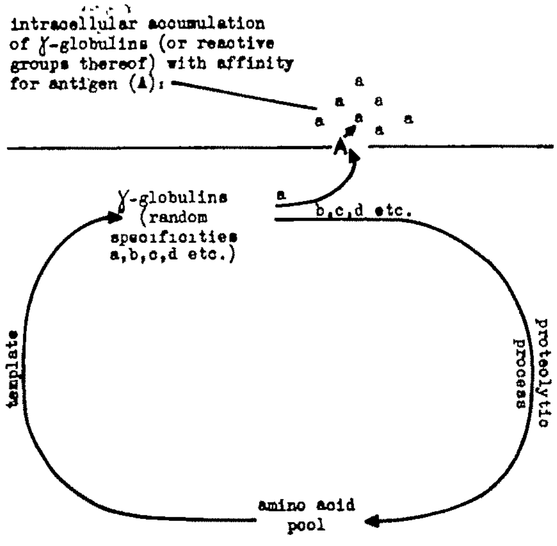
intracellular accumulation of γ-globulins (or reactive groups thereof) with affinity for antigen (A):
a
A
γ-globulins (random specificities a,b,c,d etc.)
a
b,c,d etc.
template
proteolytic process
amino acid pool

Fig. 1

matures. This, of course, would be expected to apply also to foreign molecules if they happen to be present in the body before maturation of the immune mechanism (for example, at birth) and as long as they persist in high enough concentration to neutralize all the complementary receptors which are produced.

In connexion with the actual production of specific antibodies by the antibody-forming cell after it has adsorbed antigen, we make the following assumptions. In the first place, this specialized type of cell is capable of producing γ-globulin molecules of random specificities at an extremely high rate. Secondly, the cell is capable of breaking down these molecules at about the same rate as they are synthesized. In other words, there is a 'globulin cycle', consisting first of the synthesis from an amino-acid pool of γ-globulins of different specificities, and then of their catabolism by a proteolytic system to the constituent amino-acids.

It is assumed that the antigen, after combining with the 'antigen receptor', enters the cell and that the whole molecule (or determinant groups from it) becomes fixed to some cell component in such a way that it is protected from enzymatic breakdown, rather as has been postulated for the penicillin molecule in penicillinase adaptation in *B. cereus*[19]. The complex, consisting of antigen (or determinant group) and a cell component, is placed within the cell in close proximity to the site of the γ-globulin cycle of synthesis and breakdown. In this situation it interferes with the cycle by drawing aside any molecules which have an affinity for it, passing them on to a storage site elsewhere in the cell (Fig. 1). There is thus a competition between the proteolytic system and the antigen (or determinant group) for all molecules which tend to have an affinity for the antigen (or determinant group). Molecules with no affinity for the antigen will, of course, be broken down as previously.

Thus it is postulated that the presence in the cell of the antigen (or determinant group) is soon followed by the intracellular accumulation of γ-globulin molecules (or perhaps reactive pieces of γ-globulin molecules[20]) possessing an affinity for it. Once this accumulation has occurred, the cell itself must be regarded as 'modified'; perhaps some of the antibody molecules find their way to the cell surface, rendering the cell capable of taking up many more molecules of the same antigen. The consequent increase in uptake of antigen results in further accumulation of antibodies. The metabolic effects of these intracellular changes may be expected to stimulate cell division, so that, in the presence of antigen, 'modified' cells tend to multiply at a greater rate than 'normal' cells of the same series, resulting in the formation of clones of antibody-forming cells. Sooner or later most of the specific globulins, or antibodies, are released into the circulation.

It could be argued, on teleological grounds, that such a process would seem unlikely because it involves an enormous degree of wastage in the synthesis of γ-globulin. However, there are, of course, numerous examples in Nature of apparently wasteful phenomena. Moreover, in this instance there is no actual loss of building material, since the amino-acids are returned to the pool to be used again for further synthesis of γ-globulins, and the 'wastage' applies only to the energy used in the 'globulin cycle'. On the other hand, it is not essential to the main hypothesis to assume complete breakdown of the γ-globulin molecules. One could also suppose, for example, that only the last stage of the process, namely the folding, is reversible and that there exists a pool of γ-globulin molecules under conditions which allow a constant oscillation between the folded and the unfolded state, so that molecules with new affinities are continuously being formed. This possibility comes close to Pauling's theory[8], but differs from it in that it implies a selective rather than an instructive role of the antigen's determinant group.

Since this hypothesis does not require that the antigen modifies, either directly or indirectly, the structure of the γ-globulin molecules synthesized, it qualifies for inclusion in the category of 'elective', rather than 'instructive' antibody theories [21,22]. However, unlike the 'clonal selection theory'[21-25], which is also in the elective group, it does not assume any nuclear or somatic mutation. It does postulate a selection of certain cells, but it is a selection of cells which have become modified as a result of contact with antigen, and not, as in the clonal theory, a selection of genetic variants. (An essential difference between a mutant and the 'modified' cell is, of course, that the former would retain its capacity to be stimulated by antigen even if it continued to multiply for many generations in the absence of the antigen; the modified cell would not.) In this respect the possibilities discussed here are similar to the ideas put forward by Monod[9] and Pappenheimer *et al.*[26], who compare antibody formation with adaptive enzyme production in bacteria. They suggest that the antigen may be captured by a specific 'permease' on the cell surface, and that afterwards more of the same permease is produced so that the capacity of the cell for taking up the specific antigen is greatly increased. The increased uptake of the antigen results in further production of permease and of antibody, and also stimulates cell division.

According to the clonal selection theory, there exist in the mammalian body a vast number of different clones of antibody-forming cells, each producing its own special γ-globulin. When antigen enters the circulation it is thought to attach itself to the cells which have reactive sites corresponding to the determinant groups of the antigen, this resulting in a selective advantage for these particular cells. The consequent shift in cell population is reflected by an increase in the circulation of γ-globulins of the type produced by the selected cells. It is suggested that cell variants possessing reactive groups for self-components are eliminated either in the embryo[23] or before they reach maturity in the adult[21,22]. Since each cell is supposed to possess reactive sites for only one determinant group (or perhaps a few determinant groups), the elimination of all immature cells with self-reactive sites would mean the destruction of only a small proportion of the total number.

For several reasons the clonal selection theory, in contrast to the hypothesis put forward above, seems to imply that the mechanism by which the antibody-forming system recognizes foreign matter is in principle different from that by which the phagocytes of primitive Metazoa distinguish between 'self' and 'not-self' components. For one thing, the number of phagocytes in these minute animals is relatively very small, so that the chances for variation must be severely limited. Moreover, each phagocyte seems to have an affinity, not for just one determinant group, but for an almost infinite variety of foreign molecules. Considering the large number of reactive sites which this would entail per cell, the chances of a given immature variant arising devoid of reactive sites for self components would seem to be extremely small. It follows that elimination of all cells with self-affinities would mean destruction of the majority, rather than the minority, of the immature variants. In this instance it is surely easier to envisage elimination of the self-reactive groups themselves, rather than of the variants possessing such groups.

The hypothesis advanced here does not have any special advantage over most of the recent theories of antibody production ; like them, it is neither proved by, nor does it conflict with, the known facts. It is hoped, however, that it succeeds in emphasizing the need for more comparative studies, especially on very primitive metazoa. Metchnikoff wrote "... l'étude de ces animaux (inférieurs), que nous présentent des conditions bien plus simples et plus primitives que l'homme et les vertebrés, nous fournit pour ainsi dire la clef des phénomènes pathologiques compliqués"[27]. Although antibody production is not itself a pathological phenomenon, these words are nevertheless very much to the point ; in relation to the problem of specific acquired immunity they are as meaningful to-day as in 1892 when they were written.

[1] Ehrlich, P., *Deutsch. med. Wschr.*, **24**, 597 (1898).

[2] Ehrlich, P., *Proc. Roy. Soc.*, B, **66**, 424 (1900).

[3] Jerne, N. K., *Proc. U.S. Nat. Acad. Sci.*, **41**, 849 (1955).

[4] Talmage, D. W., *Science*, **129**, 1643 (1959).

[5] Breinl, F., and Haurowitz, F., *Z. physiol. Chem.*, **192**, 45 (1930).

[6] Pauling, L., *J. Amer. Chem. Soc.*, **62**, 2643 (1940).

[7] Burnet, F. M., and Fenner, F., "The Production of Antibodies" (Macmillan and Co., Ltd., Melbourne, Australia, 1949).

[8] Grabar, P., Abstracts of Communications. First Intern. Cong. Biochem., 446 (Cambridge, 1949).

[9] Monod, J., in "Cellular and Humoral Aspects of the Hypersensitive States", ed. H. S. Lawrence, 623 (Hoeber-Harper, New York, 1959).

[10] Tyler, A., *Physiol. Revs.*, **28**, 180 (1948).

[11] Spiegel, M., *Ann. N.Y. Acad. Sci.*, **60**, 1056 (1955).

[12] Tyler, A., *Growth*, **10** (Supp.), 7 (1947).

[13] Weiss, P., *Yale J. Biol. Med.*, **19**, 235 (1947).

[14] Weiss, P., *Quart. Rev. Biol.*, **25**, 177 (1950).

[15] Ray, D. L., *J. Exp. Zool.*, **118**, 443 (1951).

[16] Humphrey, J. H., and Mota, I., *Immunol.*, **2**, 19 (1959).

[17] Boyden, S. V., Sorkin, E., and Sparck, J. V., Symposium on the Mechanism of Antibody Formation, Liblice, Czechoslovakia, 1959 (in the press).

[18] Boyden, S. V., and Sorkin, E. (submitted for publication).

[19] Pollock, M. R., in "Adaptation in Micro-Organisms", Third Symposium of the Society for General Microbiology (Cambridge University Press, 1953).

[20] Porter, R. R., *Nature*, **182**, 670 (1958).

[21] Lederberg, J., *J. Cell. Comp. Physiol.*, **52**, Supp. 1, 398 (1958).

[22] Lederberg, J., *Science*, **129**, 1649 (1959).

[23] Burnet, F. M., *Austral. J. Sci.*, **20**, 66 (1957).

[24] Burnet, F. M., "The Clonal Selection Theory of Acquired Immunity" (Cambridge University Press, 1959).

[25] Talmage, D. W., "Ann. Rev. Med.", **8**, 239 (1957).

[26] Pappenheimer, jun., A. M., Scharff, M., and Uhr, J. W., in "Mechanisms of Hypersensitivity", eds. J. H. Shaffer, G. A. LoGrippo and M. W. Chase, 417 (Little, Brown and Co., Boston, 1959).

[27] Metchnikoff, E., "Pathologie Comparée de L'Inflammation" (G. Masson, Paris, 1892).

VARIATION OF UPPER-ATMOSPHERE DENSITY WITH LATITUDE AND SEASON : FURTHER EVIDENCE FROM SATELLITE ORBITS

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In a recent article in *Nature*[1], it was explained how the variations in air density at heights and latitudes near that of the perigee of an Earth satellite could be found from the rate of contraction of the orbit of the satellite. The method was applied to *Sputnik* 3 (1958δ2), and it was concluded that, for latitudes between 50° N. and 65° S., the air density at a height of 226 km. did not depart from its average value by a factor of more than 1·5 as a result of variations with latitude and season, and all other causes, such as solar disturbances[2-6] (which give rise to an oscillation with an amplitude of about ± 20 per cent) and a possible, much smaller, day-to-night effect. This conclusion conflicts with earlier direct measurements from rockets[7,8], which suggested a much wider variation with latitude and season, by a factor of 5 or even 10, at heights of 200–300 km. Since the subject remains rather controversial, it seems worth giving further results which have recently become available.

The latitude-range covered by *Sputnik* 3 (50° N.–65° S.) can be extended with the aid of *Discoverer* 6 (1959ζ), the orbit of which was inclined at 84° to the equator. *Discoverer* 6 was in orbit for two months, from August 19 to October 20, 1959, and during this time its perigee moved northward from a latitude of about 50° N. to 84° N., and then southward to about 50° S. The air density at the initial perigee height of 220 km., obtained by the method previously described[1], and expressed as a multiple σ of the average density, is plotted in Fig. 1. The points in Fig. 1 are at intervals of 6 days, during which time

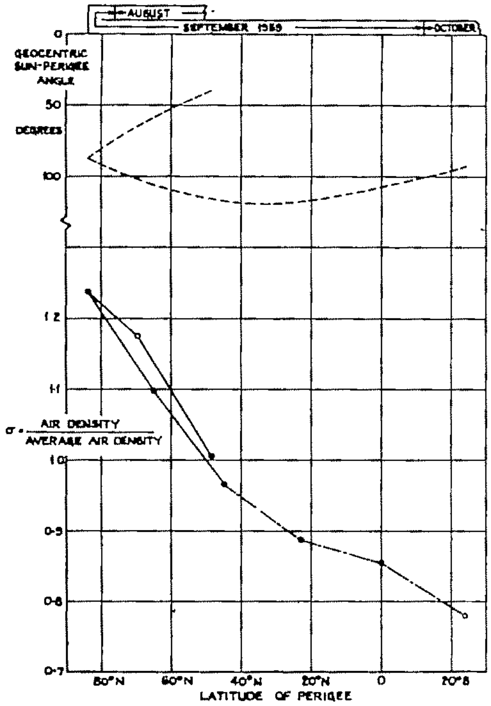

Fig. 1. Air density at a height of 220 km., deduced from the orbit of *Discoverer* 6: variation with latitude and time

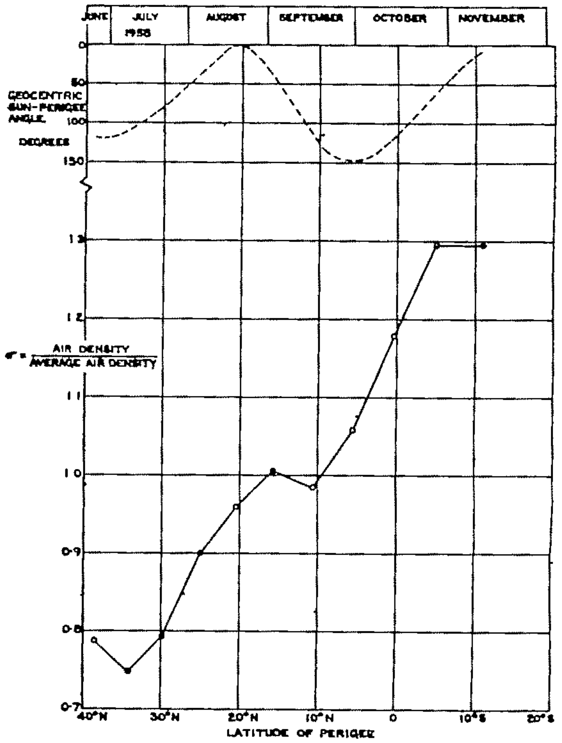

Fig. 2. Air density at a height of 220 km., deduced from the orbit of *Sputnik* 3 rocket: variation with latitude and time

the perigee latitude changes by about 20°. The main orbital data—the values of orbital period—were obtained partly from information issued by Project Space Track, Bedford, Mass., and partly from observations by the authors. Errors in the values plotted in Fig. 1 may arise from (1) errors in the orbital data, (2) errors in the assumed value for atmospheric scale height[1], and (3) changes in the mode of rotation of the satellite. The estimated standard deviation in Fig. 1 from these causes is a little more than 10 per cent.

Similar calculations have been made for the rocket of *Sputnik* 3 (1958δ_1), and the results are plotted in Fig. 2. This satellite[4] revealed strong and regular variations in air density, with amplitude about ±20 per cent and having maxima at intervals of about 28 days and minima at similar intervals. The points in Fig. 2 were calculated at intervals of 14 days, halfway between a maximum and a minimum, to smooth out these oscillations. The standard deviation in Fig. 2 is likely to be about 10 per cent, and again the relevant perigee height is 220 km.

The graphs in Figs. 1 and 2 represent, in effect, an average density over the range of latitudes near perigee where the air drag is important. For *Sputnik* 3 rocket the initial orbital eccentricity was near 0·1, and 80 per cent of the total drag effect occurred at latitudes within 15° of perigee. But *Discoverer* 6 had a lower initial eccentricity, 0·047, and the drag effect is therefore spread over a wider range of latitude, 80 per cent of it occurring at latitudes within 30 degrees of perigee. Towards the end of the life

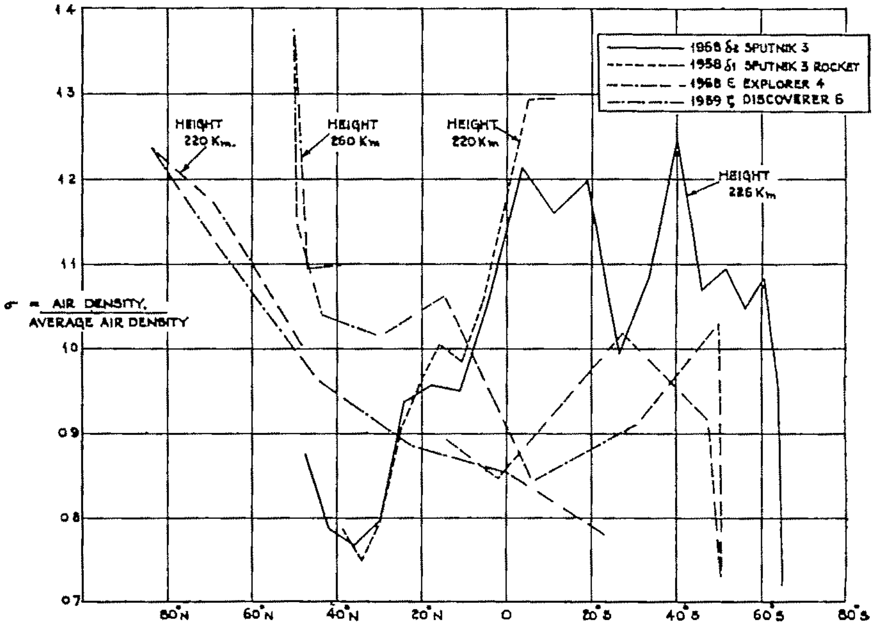

Fig. 3. Variation of air density with latitude at heights of 220–260 km, deduced from satellite orbits

of this satellite the spread in latitude is even greater, and the graph in Fig. 1 has been drawn as a broken line, since it does not represent realistically the detailed variation of density with latitude, though it does serve to indicate the average density in the equatorial region, for comparison with the density in north polar regions on the left of the graph.

Figs. 1 and 2 both confirm the earlier evidence that density does not depart from its average value, at heights near 220 km., by a factor of more than 1·5, and suggest that the variation due to latitude and season is probably a good deal smaller than this.

The angle *SCP* subtended at the centre of the Earth *C* by the sun *S* and perigee *P* of the satellite is also plotted in Figs. 1 and 2. If *SCP* exceeds 90°, the Earth below perigee is in darkness; if *SCP* exceeds about 108°, the perigee point itself is in darkness. The slight suggestion of a small variation in density between day and night indicated[1] by *Sputnik* 3 is given little support by Figs. 1 and 2. At heights around 220 km. it seems probable that day-to-night changes in density do not exceed ± 10 per cent.

The values of density at heights of about 220 km. given by *Sputnik* 3 (ref. 1), *Discoverer* 6 (Fig. 1) and *Sputnik* 3 rocket (Fig. 2) are plotted against latitude in Fig. 3, together with similar results for heights near 260 km. obtained from *Explorer* 4 (1958ε) by Schilling and Whitney[9] (though the method of presentation of these latter results has been altered, to fit in with the scheme of Fig. 3). The first point of interest in Fig. 3 is the close agreement between the results from *Sputnik* 3 and its rocket. This is to be expected because the perigees of the two satellites reached a given latitude at roughly the same dates and were therefore sampling the same regions of air. The small difference, averaging about 4 per cent, between the two sets of values suggests that the error inherent in the method of calculation is less than the 10 per cent mentioned earlier.

Apart from this close correspondence between the results from *Sputnik* 3 and its rocket, Fig. 3 is notable for an almost complete absence of significant structure: the only positive conclusion which might possibly be drawn is that density tends to be rather high north of lat. 50° N. and rather low south of lat. 60° S. On the whole, however, it appears more likely that the variations are unconnected with latitude. When the values of σ are plotted against the difference between perigee latitude and the declination of the Sun, to indicate possible seasonal effects, the resultant graph, which is not reproduced here, resembles Fig. 3 and again shows no sign of significant structure. The obvious conclusion is that the variations in density are controlled by time-dependent sources, for example, solar disturbances, rather than by latitude and season, which depend essentially on the position of the satellite relative to the centre of the Earth.

To sum up the results of this and previous work, it appears that solar disturbances are the main factor influencing the air density at heights of 200–300 km., giving rise to variations which have an amplitude of about ± 20 per cent and a periodicity of about 28 days. Day-to-night changes may also possibly have an effect, with the density not more than 10 per cent above average by day. Variations with latitude and season are small, and, when density is plotted against latitude or season, no consistent pattern emerges. At greater heights, 300–700 km., the variation from day to night is much more important[10,11] and has a greater amplitude than the 28-day variation due to solar disturbances.

[1] King-Hele, D. G., *Nature*, **184**, 1267 (1959).
[2] Nonweiler, T. R., *Nature*, **182**, 468 (1958).
[3] Jacchia, L. G., *Nature*, **183**, 526 (1959).
[4] King-Hele, D. G., and Walker, D. M. C., *Nature*, **183**, 527 (1959).
[5] Priester, W., *Naturwiss.*, **46**, 197 (1959).
[6] Jacchia, L. G., *Nature*, **183**, 1662 (1959).
[7] Kellogg, W. W., *Planet. Space Sci.*, 1, 71 (1959).
[8] LaGow, H. E., Horowitz, R., and Ainsworth, J., paper presented at Fifth CSAGI Assembly (Moscow, 1958).
[9] Schilling, G. F., and Whitney, C. A., *Planet. Space Sci.*, 1, 136 (1959).
[10] Jacchia, L. G., Smithsonian Astrophys. Obs. Spec. Rep., 29 (1959).
[11] Martin, H. A., and Priester, W., *Nature*, **185**, 600 (1960).

THE APPROACHING DESCENT OF *SPUTNIK* 3 (1958δ2)

By B. R. MAY

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SINCE *Sputnik* 3 (1958δ2) is nearing the end of its life-time, it may be of some interest to give a short account of its progress since the last review[1], and to discuss the likelihood of favourable observing periods from the British Isles, before its descent.

Throughout May, most of July, and August, the satellite was illuminated by the Sun on at least one transit each night, and on some nights as many as four consecutive transits were visible from the British Isles. This was due to the high declination of the Sun and the comparatively great heights (925 km., and above) at which the satellite traversed our latitudes. The fine weather enabled visual observers to obtain regular sightings, at stellar magnitudes ranging from + 3 to + 9. During June, however, the near transits occurred in the day-time, so that only radio observations were possible during that month. Since about the end of August the transits have been subject to varying eclipse conditions, and the number of visual observations made has generally decreased due to this, and to a greater extent, to the unfavourable weather conditions. Because of the rotation of the perigee of this orbit towards the northern hemisphere, the height of transit over our latitudes has been steadily decreasing since the end of June 1959, when apogee was at the maximum northern latitude. At the time of writing (February 15), the height of the satellite over our latitudes ranges from 220 to 470 km.

Since the perigee distance of this satellite is fairly small—about 6,560 km.—the magnitude of the atmospheric drag experienced by the satellite at its perigee will change appreciably as the perigee rotates over the oblate Earth. Thus, the estimation of the life-time of the satellite, which is based largely upon the drag, is subject to uncertainties even after the orbit is well known. In May 1959 a theoretical date of descent in early January 1960 was indicated—the present estimate places it in late March 1960.

For most of the few remaining weeks of the life of the satellite the near transits will take place in the day-time and, since the transmitters of the satellite are powered by solar cells only, good opportunities for radio tracking will be afforded. Transits for visual observations from the British Isles will occur from about March 16 onwards, in the mornings. Due to its low height, *Sputnik* 3 may well become a comparatively bright object, and it is hoped that this will increase the chances of valuable observations being made upon the last revolutions of this most interesting and successful satellite.

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[1] May, B. R., and Smith, D. E., *Nature*, **184**, 765 (1959).

THE CANADIAN COMMITTEE ON OCEANOGRAPHY

THE Canadian Committee on Oceanography represents the re-organized Canadian Joint Committee on Oceanography which came into existence on April 1, 1946. It is comprised of representatives of the following Government agencies: Royal Canadian Navy, Fisheries Research Board, Department of Mines and Technical Surveys, Defence Research Board, National Research Council, Meteorological Branch of the Department of Transport, and Marine Services of the Department of Transport. In addition there are representatives of academic interests in oceanography nominated by the National Research Council.

The Committee is under the chairmanship of Dr. W. E. van Steenburgh, director general of Scientific Services, Department of Mines and Technical Surveys, Ottawa, Ontario. The secretary is Dr. H. B. Hachey, chief oceanographer, Fisheries Research Board of Canada, Ottawa, Ontario.

The functions of the Committee are to: (*a*) coordinate the oceanographic activities of all Canadian Government agencies; (*b*) act, as designated by the National Research Council, as the national committee for the Scientific Committee on Oceanographic Research; (*c*) act, as designated by the National Research Council, as the Section on Oceanography of the Council's Associate Committee on Geodesy and Geophysics.

Each of the co-operating organizations is either actively engaged in oceanographic investigations or contributes in ships, laboratory facilities, personnel or funds towards the Canadian programme, the total effort being co-ordinated through the Committee.

Activities coming under the Canadian Committee can be divided according to regions under investigation.

(*a*) *Atlantic coast.* This includes general surveys of the Gulf of St. Lawrence, the Scotian Shelf, the Bay of Fundy region, Newfoundland waters, and the Labrador and sub-Arctic seas; studies on slope waters and the mechanism of the Gulf Stream system, on the fundamentals of the Gaspe Current and Cabot Stream, on tides and currents, on cyclic variations in water temperatures, on problems of the varying influence of Atlantic and Arctic waters, on the formation and movements of ice, on biological, geological and geophysical problems, and special studies involving fisheries, naval and other national problems.

(*b*) *Pacific coast.* The work here covers general surveys of the waters of the eastern North Pacific, the Straits of Georgia and Juan de Fuca, Hecate Strait, and the fjord-like British Columbia inlets; studies on pollution in the sea as related to pulp-mill effluents and sewage; studies of current patterns and tides in Nature, and investigations of the fundamentals of flow by means of models; productivity studies, and variations in water characteristics as followed by observations at a network of coastal stations and from the Canadian Weathership; studies on biological, geological and geophysical problems and special studies involving fisheries, naval, and other national problems.

(*c*) *Arctic coast.* General surveys are being made of the waters of the Canadian Arctic, extending from the Bering Strait and Beaufort Sea to Baffin Bay and Davis Strait, with particular attention to the water characteristics in the various sub-areas and the volume and direction of flow through the various passages. An extensive survey is being undertaken of the Polar Continental Shelf, involving general oceanographic observations, tidal and current studies, geological, geophysical and biological investigations.

The work of the Committee is being carried out at a number of institutions. There is an Institute of Oceanography at the University of British Columbia, Vancouver, B.C., under the directorship of Dr. George Pickard, and an Institute of Oceanography at Dalhousie University, Halifax, N.S., has recently been set up under the directorship of Dr. F. R. Hayes.

The Pacific Naval Laboratory at Esquimalt, B.C., and the Naval Research Establishment at Dartmouth, N.S., operated by the Defence Research Board, conduct oceanographic research and surveys in support of military requirements.

Plans for the Bedford Institute of Oceanography, near Halifax, N.S., have been announced and the development is now under way. The Institute, which is to be completed in five years, will have a staff of 300 oceanographers, hydrographers, submarine geologists and other scientific personnel, plus supporting staff, and an operating fleet of ten oceanographic vessels. The Atlantic Oceanographic Group of the Fisheries Research Board will also be located in the Institute. This Institute will be under the Department of Mines and Technical Surveys. In support of its work, a substantial shipbuilding programme is planned. The first ship, the C.G.S. *Hudson*, which is expected to cost 7 million dollars, is to be commissioned in 1961. Dr. W. M. Cameron has recently been appointed to take charge of the development of the Bedford Institute. Widely known for his work in oceanographic research, Dr. Cameron has made special studies in the fields of biological and anti-submarine research. He is best known, however, for his Arctic studies and for his work in theoretical oceanography, particularly the theory of circulation in estuaries. His Arctic studies were mainly carried out as a member of the joint Canada–United States Beaufort Sea Expeditions, in which he was senior Canadian scientist. He was for a time professor of oceanography in the University of British Columbia, where he established an Institute of Oceanography. Since 1958 he has been director of plans of the Defence Research Board in Canada.

BIRDS AND FORESTS

THE Council of the British Trust for Ornithology has recently taken the initiative in promoting discussions with other bodies interested in economic aspects of ornithology. The second of its series of joint conferences was hold at Northerwood House, Lyndhurst, Hampshire, during October 30–November 1. The Forestry Commission was the Trust's partner in organizing the conference, and the subject was "Birds and Forests".

In opening the conference, Mr. E. M. Nicholson (Nature Conservancy) reviewed the relations between birds and forestry. He pointed out that remarkably little experimental work had so far been done in Britain to assess the part played by birds in the forest economy; the work of the Edward Grey Institute of Field Ornithology on tits and other birds in the Breckland and the Forest of Dean, and of H. N. Southern, of the Bureau of Animal Population, on the tawny owl in Wytham Wood, were examples of what could be done. He called for a new look at the problems of forest ornithology as a whole.

Mr. T. R. Peace (Forestry Commission) referred to the recent shift of emphasis from the idea of creating a strategic reserve of timber to that of economic forestry. This meant turning away from the natural forest towards a greater degree of deliberate tree farming. The Commission, as its creation of National Forest Parks has shown, was always prepared to pay attention to amenity; but economic considerations do not permit forests to be designed primarily for the encouragement of wild life. The Forestry Commission had not so far undertaken any research on problems of forest ornithology because it did not believe a case had yet been made out for birds being sufficiently beneficial to woodlands under British conditions to justify this.

In the subsequent discussion the point was brought out that the forester was faced with a choice between the small but continuing expense of providing and maintaining nest-boxes in order to increase the population of pest-predator birds and the much greater, but only very occasional, expense of attacking an infestation with sprays.

Mr. A. R. Mitchell (Forestry Commission) described the impact of forestry practice on bird life in woodlands. Wood-larks, for example, found forest nurseries a suitable habitat. On the other hand, the first thinning of a new plantation removed the forked and crooked trees in which birds liked to nest.

Dr. J. A. Gibb (Animal Ecology Section, Department of Scientific and Industrial Research, New Zealand; formerly of the Edward Grey Institute of Field Ornithology, Oxford) sent a paper describing the work of the Breckland Research Unit of the Edward Grey Institute with special reference to populations of insectivorous birds and their food supply in pine plantations. The Unit was able to demonstrate that the density of tits and goldcrests in the Breckland pine plantations (mainly 20–30 years old) in winter varied closely with their stock of food. In severe winters, the tits and goldcrests might be actually consuming 40 per cent of the total amount of insect food available for them, and very much larger proportions of the available supplies of their favourite foods. Though this did not by itself prove that the birds' numbers were being controlled in a density-dependent way by food shortage, it did prove the existence of the indispensable condition needed for this method of control to operate.

The result of the Breckland work was to suggest that the provision of nest-boxes, though increasing the local density of birds in the breeding season, could not enable a greater number of tits to survive the crucial winter season in any particular forest. It might indeed be more to the point to increase the stock of winter food available for the birds by planting suitable trees and shrubs on the forest margins to diversify the supply; for example, spruce and beech would provide autumn stocks of food for coal tits, which they could store for use in the winter (pines shed their seeds in spring).

As regards the impact of birds on insect populations, the Breckland researches showed that bird predation accounted for a substantial proportion of insect stocks, so that birds are a force to be reckoned with in forest economics.

Dr. Herbert Bruns (Staatliche Vogelschutzwarte, Hamburg) described various aspects of forest research on economic ornithology in Germany. Food studies showed that, on the whole, insect-eating birds fed chiefly on pest-insects, sometimes in large quantities. This did not, however, necessarily prove that they were important controls on these insects. However, there were several convincing examples in Germany where it could be proved that birds had actively controlled insect pests. In one pine forest in Baden it had been shown that insect infestation was much worse in nearly birdless monocultural plantations than in bird-rich woods nearby. When more nest-boxes were brought into the wood, the infestation was reduced.

Summing up, Dr. Bruns said that birds were not a panacea for all forest ills; but an important link in the natural chain of factors making for a healthy forest. Methods of sylviculture were most important, and it must be borne in mind that most German forests were much more mature than the average British forest. Much of the different British experience might be due to the immaturity of many British forests. The critical age for pest-devastation came at 60–70 years.

Mr. R. K. Murton (Ministry of Agriculture) discussed the control of wood-pigeons, which might be a serious forest pest, especially to the leading shoots of Sitka spruce and other species. In East Anglia at least the Forestry Commission could be absolved from generating a high population of wood-pigeons, for their plantations were too extensive. Dense nesting of wood-pigeons only occurs in a fringe about 100 yards deep around the edges of large blocks of woodland, since the birds are unwilling to nest very far from their feeding-grounds. Wood-pigeons therefore prefer to breed in hedgerows, shelter-belts and the small spinneys planted as shelter for game. The game preserver indeed was largely responsible for the existence of the plague of wood-pigeons in East Anglia, for in addition to providing their nesting places, he prevented any control measures until February, after the shooting season, when much damage had already been done to the crops of farmers and foresters. Moreover, it was the keeper who killed off the two principal predators of the wood-pigeon, the jay and the magpie, which are responsible for 80 per cent of the losses of wood-pigeons' eggs.

Dr. Bruce Campbell (British Trust for Ornithology) discussed population problems in a typical insect-eating forest bird, the pied flycatcher, illustrated by the ringing and nest-box experiments he was undertaking in the Forest of Dean.

Mr. W. A. Cadman (Forestry Commission) described the progression of wild life at various stages in the development of forests. He said that the pine marten had increased in some Welsh forests.

Mr. Nigel Gray (Imperial Chemical Industries Game Research Station) discussed the relations between game preservation and forestry. He said that good forestry made for good game habitat; but it was essential to have a reasonable mixture of tree species. If the Forestry Commission were to appoint game advisers, it would succeed in greatly boosting private forestry, by showing owners ho it could be combined with game preservation.

Dr. I. D. Pennie (British Trust for Ornitholog spoke about the capercaillie and its effects on woo lands. Foresters claimed that in autumn and wint the capercaillie ate almost exclusively the shoots an buds of conifers; but in fact it seemed much t prefer the shoots of transplanted trees in nurseri and plantations to those of naturally regeneratin trees. It was thus the shift in forestry practice, fro the natural woodland to the artificial plantation, tha had transformed this normal member of the woodlan avifauna of northern Europe into a forest pest i parts of Scotland.

There were two field excursions during the conference, one under Mr. B. Gale, with a forestry angl and one under Mr. Edwin Cohen, mainly for bird watchers.

R. S. R. FITTER

OBITUARIES

Mr. E. Price Evans

MR. EVAN PRICE EVANS died on April 27, 1959, at his home in Eastbourne, aged seventy-seven. He will be remembered for the part he played in introducing the teaching of plant ecology into schools; it is probably quite largely due to him that at least in some schools ecological teaching on scientific lines has taken the place of mere 'Nature study'.

Price Evans was a Welsh-speaking Welshman, born at Corris in Merioneth on January 19, 1882. Educated in his native village and at Towyn County School, he entered the University College of North Wales, Bangor, in 1900 and was trained to be a teacher. There he attended the botanical lectures of Prof. Reginald Phillips and Dr. (later Prof.) J. Lloyd Williams. His first appointment was at his own school at Towyn, under the headmastership of a notable chemist, Thomas Jones.

At Towyn he had already experimented with teaching field botany even to quite junior forms, and he later developed this teaching when holding a post at Ryhope, Co. Durham. In his youth, however, he was better known as a footballer than as a scientist and at one time played for Wales in the amateur internationals. From Ryhope he went to Warrington, where he became headmaster of the Grammar School.

At Ryhope from 1913 onwards he began teaching ecology and vegetation mapping to his classes, and his efforts in this direction led him to correspond with the late Sir Arthur Tansley, who was then at Cambridge. Tansley encouraged him to publish an account of his teaching methods in the *School Science Review*, and later they wrote in collaboration "Plant Ecology and the School" (1946).

Price Evans was a keen student of vegetation on his own account, especially that of his native mountains, which he knew very intimately. He contributed several papers to the *Journal of Ecology*, of which one dealing with Cader Idris is still the only published account of the vegetation of a geologically and botanically very interesting area and is often quoted. He was an accurate observer and kept notes methodically; his unpublished note-books are full of valuable observations on the plants and plant communities of North Wales. The University of Wale recognized his work by awarding him the honorar degree of M.Sc. in 1933.

During and after the Second World War, Price Evans was a keen supporter of Nature conservation and he was an active member of the Nature Conservancy's Committee for Wales from its inceptio until shortly before his death. He had a vigorou and friendly personality and will be much missed.

P. W. RICHARDS

Dr. Z. K. A. Moszynski

DR. Z. K. A. MOSZYNSKI, a principal scientifi officer of the British Coke Research Association died at his home in Chesterfield, Derbyshire, on February 13. Zbigniew Konrad Antoni Moszynsk was born at Lwow, Poland, on December 16, 1907 and studied in the Department of Chemical Engineering, University of Lwow (1927–34), where he took his diploma (Dipl. Ing.). During 1934–35 he worked in the Laboratory of Potassium Salts, University of Lwow. During 1935–39 he was deputy inspector for civil defence of industry, Polish Ministry of Defence in the County of Lwow.

In August 1939 he was mobilized and served in the Polish–German campaign, escaping to Hungary where he was interned. For five months he was assistant to the professor of agricultural chemistry University of Magyarovar, but in March 1940 he joined the Polish forces in France, being evacuated to England. On behalf of the Polish General Staff he carried out research work in the Mining Department University of Birmingham, between 1944 and 1945 on the use of carbonized peat/coal blends for mobile gas generators. During 1946–47 he continued and extended this work on the production of reactive carbonized fuels in the Fuel Department, University of Leeds, and obtained the degree of doctor of philosophy.

In September 1947 he was demobilized and in the same year became a British subject. He joined the staff of the British Coke Research Association, at the Midland Coke Research Station, where he was in charge of personnel and acted as secretary of a research group working on the analysis of coal and coke. His bomb combustion method for the determination of both sulphur and chlorine in coal is well known to fuel scientists, and is to become an international standard method. With the building of the new Coke Research Centre at Chesterfield in 1958 he transferred his home to Derbyshire and took up new work for the British Coke Research Association dealing with the problems of air and water pollution.

NEWS and VIEWS

Physiology at Oxford :

Prof. E. G. T. Liddell, F.R.S.

The near prospect of Prof. E. G. T. Liddell's retirement from Oxford's Waynflete professorship of physiology has aroused a widespread interest among physiologists everywhere and, indeed, among all those who are concerned for the advancement by experimental research of knowledge in the medical range of the sciences. For there can be no doubt that the Waynflete chair, during the tenures of a succession of distinguished occupants, has achieved and maintained a reputation second to that of no other chair of physiology, anywhere in the world. Sir John Burdon-Sanderson and his pupil and successor, Prof. Francis Gotch, made contributions of fundamental importance to knowledge of the nature of nervous impulses, as excited and propagated in the continuity of nerve fibres. Under Sir Charles Sherrington the reputation of the Waynflete chair, and of the Oxford School of Physiology attached to it, attained additional eminence, through the discoveries which he and a series of distinguished associates made concerning the intricate phenomena of the central nervous system, involving the transmission of nervous excitations across systems of synaptic junctions of varying complexity; discoveries which have provided a large component of the foundation for modern neurological theory and practice. It is in this field of research, on the physiological processes of the brain and the spinal cord, that Prof. Liddell, as an intimate pupil of and collaborator with his great predecessor, has maintained that distinguished tradition.

Sir Lindor Brown, C.B.E., F.R.S.

Under Sir George Lindor Brown, Jodrell professor of physiology in University College, London, whose appointment to succeed Prof. Liddell in the Waynflete chair has recently been announced, it may be confidently expected that the tradition will be further maintained. After a distinguished student career in physiology at Manchester, Brown held a lectureship at Leeds, where the late B. A. McSwiney was then the professor. Work there, with McSwiney and others, gave early evidence of Brown's interest in the special physiology of the autonomic nervous system, and in the then recent evidence for a chemical mechanism of excitatory transmission at its ganglionic and peripheral junctions. With a grant from the Medical Research Council, he also worked for a period in Sherrington's Oxford department with J. C. Eccles. He then, in 1934, accepted an invitation to join Sir Henry Dale's team at the National Institute for Medical Research, and played a prominent part in extending the evidence for a cholinergic mechanism, to the transmission from motor nerve endings to the end-plates of voluntary muscle. Brown continued investigations in this general field with a series of distinguished colleagues and visiting experts after Sir Henry's retirement in 1942, having meanwhile become engaged in important war-time researches and responsibilities. In 1949 he became professor of physiology in University College, London, where another famous chair of physiology will be vacated by his removal to Oxford; and since 1955 he has been the biological secretary of the Royal Society. The chief focus of research-interest in the Oxford School of Physiology may, perhaps, move for a time from the central towards the peripheral nervous system; but there need be no fear of a lowering of its standard.

Astronomy at Oxford : Prof. H. H. Plaskett, F.R.S.

On September 30, Prof. H. H. Plaskett retires under the age limit, after twenty-eight years as Savilian professor of astronomy and director of the University Observatory at Oxford. His tenure of office has been notable for the building up of a school of solar physics, now well known for the thoroughness of its methods, the clarity of exposition in its lectures, and its avoidance of popular publicity. With only modest resources, two new solar telescopes and large spectrographs have been built and operated successfully. Prof. Plaskett's own work has been both observational and theoretical. He has studied models of the solar atmosphere, the formation of Fraunhofer lines, the structure of the solar granulation and problems of the chromosphere. Under his inspiration and guidance his colleagues and pupils have also made important contributions to solar physics. Among them they have discovered the strengthening of weak Fraunhofer lines from centre to limb, they have invented elegant interferometric methods for measurement of Fraunhofer and chromospheric lines, developed improvements in the mathematics of stellar atmosphere theory, and made long series of precision measurements to try to find whether the Einstein red-shift can be detected in the solar spectrum. Finding that the red-shift is obscured by spectrum line displacements arising from other causes, they have been led on one hand to study velocity fields in the solar photosphere, and on the other to make laboratory measurements of pressure shifts in atomic spectra of astronomical importance. Astronomy could well do with more of the devotion to careful measurement and the restraint on speculation which have characterized Oxford work under Prof. Plaskett.

Prof. D. E. Blackwell

Dr. D. E. Blackwell, who is to succeed Prof. Plaskett, has been assistant director of the Solar Physics Observatory, Cambridge, since 1948. He entered Sidney Sussex College in 1943 as a major scholar, from the Merchant Taylors' School, took first-class honours in both parts of the Natural Sciences Tripos, was an Isaac Newton Student in 1946 and Stokes Student at Pembroke College in 1947. His astronomical work has dealt chiefly with the solar corona and the zodiacal light. In a series of well-planned, adventurous and successful expeditions, in the Sudan for the 1952 eclipse, in an open aircraft at 30,000 ft. for the 1954 eclipse, over the Pacific in another aircraft in 1955, in the high Andes in 1958, he has extended optical measures until we now have reliable curves of brightness and polarization from the inner corona to the zodiacal light region 70° away. With M. F. Ingham he has photographed the zodiacal light spectrum with higher resolution than hitherto attained; their results have considerably altered our views on electron densities in interplanetary space and on the light scattered by interplanetary dust. They have also shown beyond doubt that the zodiacal light is sometimes variable. In 1956, and again in 1957, Blackwell and A. Dollfus made pioneer photographs of solar granulation from a balloon at 15,000–20,000 ft., using an 11-in. telescope, and demonstrated that most of the loss of telescope definition from bad day-time seeing can be avoided in this way. Dr. Blackwell is one who believes that observational astronomers must at all times seize upon any new techniques which promise significant advantage over older methods, and has acted on this belief with notable success.

Biology at Melbourne (Monash): Prof. A. J. Marshall

Dr. A. J. Marshall, reader in zoology and comparative anatomy in the University of London at St. Bartholomew's Hospital Medical College, where he is head of the Department, has been appointed to the chair of biology in the new Monash University in Melbourne, Australia. Dr. Marshall is an Australian by birth and upbringing; soon after entering the University of Sydney he joined the Oxford University Expedition to the New Hebrides, and then went to Oxford to help work out the results before returning to Sydney to finish his course and to graduate. During 1941–45 he was engaged in military service in northern Australia and New Guinea, and commanded the independent infantry "Jockforce" in the Wewak Campaign. In 1946 he returned to Oxford, where he obtained his D.Phil. and, later, D.Sc. degrees. In 1949 he resigned his Beit Memorial Research Fellowship to accept his appointment at St. Bartholomew's.

Dr. Marshall has always been a naturalist, and in his early years published a number of papers on natural history subjects in Australian journals. Under academic influence, and the guidance of Dr. John R. Baker and the late Dr. F. H. A. Marshall, his research interests concentrated on the physiology of reproduction in vertebrates, especially in its relation to behaviour, migration, and the ecology of breeding seasons. He has been strikingly successful in combining field and laboratory work by using the changing environment as a kind of natural laboratory. Key localities and situations, such as deserts, the arctic, early springs, and unusually severe winters, have been selected, and free-living birds, fishes, reptiles and mammals have been collected at periods, often in successive years, that were expected to yield the most valuable results upon subsequent histological investigation of their sexual cycles. He has always kept one foot in the field while the other was in the laboratory. One of his most important discoveries is the post-breeding metamorphosis of the seminiferous tubules in non-mammalian vertebrates into a mass of cholesterol-positive lipoid material which contains progesterone, and the somatic and psychological effects of the action of this hitherto undescribed endocrine gland. A very long series of papers by Marshall and his colleagues record these and cognate researches, crowned by his penetrating study of reproductive physiology and behaviour of the bower birds published by the Clarendon Press in 1954. Prof. Marshall will not only build up a first-rate teaching department at Monash, but will also undoubtedly establish a school of research in field-endocrinology, for which Australia offers unbounded opportunities, that will redound both to his own credit and that of his colleagues in the Old Country who first guided his steps towards this rewarding subject of research.

Cyto-Genetics at the Royal Botanic Gardens, Kew: Dr. Keith Jones

Dr. Keith Jones, a graduate of the University College of Wales, Aberystwyth, has been appointed a principal scientific officer at the Royal Botanic Gardens, Kew, to undertake research in cyto-genetics. Since 1948 he has been in charge of the Department of Cytology at the Welsh Plant Breeding Station, Plas Gogerddan, Aberystwyth, where his investigations have been connected mainly with European species of the Gramineae, in particular those of the genera *Agrostis*, *Anthoxanthum*, *Dactylis* and *Holcus*. He was awarded a doctorate by his University in 1957 for his thesis on "Species Differentiation in *Agrostis*", and recently spent a year studying at the Plant Research Institute, Ottawa, as a postgraduate fellow of the National Research Council of Canada.

Although Kew is internationally renowned for the beauty of its Gardens, it is primarily a research establishment concerned with the classification, determination and description of plants. For more than a hundred years its vast collections of herbarium specimens supplemented by living plants have been the subject of intensive morphological and anatomical studies. On the other hand, these unrivalled living collections, estimated to include some 45,000 species and varieties, have received comparatively little attention from the cyto-geneticists, and probably half of them have never been examined cytologically. Here, with such a wealth of desirable material, constantly being enriched by new discoveries, and representative of all groups of vascular plants and of very diverse phytogeographical regions, there is a unique opportunity for extensive cytological and genetical investigations. While the urgent need at present is for karyological studies, especially the determination of numbers and forms of chromosome sets, other lines of research, such as the genetical study of species and putative hybrids impossible of satisfactory classification by ordinary taxonomic methods, are also very necessary. It is hoped that this closer association of systematic botany, cytology and genetics at Kew will lead to the resolving of many difficult taxonomic problems and ultimately to a much improved and more natural system for the flowering plants.

Royal Society of Edinburgh: New Fellows

At a meeting of the Royal Society of Edinburgh, held on March 7, the following were elected Fellows of the Society: Dr. I. R. C. Batchelor, physician superintendent, Dundee Royal Mental Hospital, and head of the Department of Psychiatry, University of St. Andrews; Dr. D. Bijl, lecturer in natural philosophy, University of St. Andrews; Prof. T. L. Cottrell, Department of Chemistry, University of

Edinburgh; Prof. R. E. Coupland, Department of Anatomy, Queen's College, Dundee; J. A. Crichton, head of the Applied Nutrition Department, Rowett Research Institute, Bucksburn, Aberdeen; Prof. K. W. Donald, Department of Medicine, University of Edinburgh; Dr. W. J. Eggeling, conservation officer for Scotland, Nature Conservancy; Prof. R. B. Fisher, Department of Biochemistry, University of Edinburgh; R. S. Glover, officer-in-charge, Scottish Marine Biological Association's Oceanographic Laboratory, Edinburgh; W. Hutton, deputy-chairman of the South of Scotland Electricity Board; T. R. M. Lawrie, district geologist in charge of Highland work, H.M. Geological Survey, Edinburgh; R. D. Lord, senior lecturer in mathematics, Royal College of Science and Technology, Glasgow; Dr. J. A. Lovern, head of the Lipids and By-products Section, Torry Research Station, Aberdeen; Dr. D. M. C. MacEwan, lecturer in physics, Queen's College, Dundee; Prof. J. L. Malcolm, Department of Physiology, University of Aberdeen; Prof. J. A. Roper, Department of Genetics, University of Sheffield; Dr. F. A. Rushworth, lecturer in natural philosophy, University of St. Andrews; Dr. J. M. Shewan, head of the Bacteriology Section, Torry Research Station, Aberdeen; Prof. M. G. P. Stoker, Department of Virology, University of Glasgow; Prof. P. D. Sturkie, Department of Poultry Physiology, Rutgers University, New Brunswick (presently S.P.S.O., Agricultural Research Council Poultry Research Centre, Edinburgh); Prof. P. A. Sweet, Department of Astronomy, University of Glasgow; Prof. W. J. Tulloch, Department of Bacteriology, Queen's College, Dundee; Dr. G. B. Warburton, senior lecturer in charge of the Postgraduate School of Applied Dynamics, University of Edinburgh; Prof. P. E. Weatherley, Department of Botany, University of Aberdeen; Dr. R. Westwater, manager, Technical Service Department, Imperial Chemical Industries, Ltd. (Nobel Division).

Royal Geographical Society Awards

H.M. THE QUEEN has approved the award of the Royal medals of the Royal Geographical Society as follows: *Patron's Medal*, to Prof. Théodore Monod, director of the Institut français d'Afrique Noire, for geographical exploration and research in the Sahara; *Founder's Medal*, to Phillip G. Law, leader of the Australian National Antarctic Research Expeditions, for Antarctic exploration and research; *Victoria Medals*, to Dr. Armando Cortesão, for studies in the history of Portuguese cartography, and Prof. J. A. Steers, University of Cambridge, for research in coastal morphology; *Murchison Grant*, to Prof. J. Wreford Watson, University of Edinburgh, for contributions to the development of geographical studies in Canada; *Back Grant*, to A. T. Grove, University of Cambridge, for geomorphological studies in Northern Africa; *Cuthbert Peek Grant*, to G. F. Reid, directorate of Overseas Surveys, for contributions to Antarctic surveying; *Gill Memorial*, to Joyce M. Lambert, University of Southampton, for research in the origin of the Broads; and *Mrs. Patrick Ness Award*, to R. J. Adie, University of Birmingham, for contributions to Antarctic geology.

Institute of Metals Awards

THE Institute of Metals has made the following awards: *Institute of Metals (Platinum) Medal* for 1960, to Prof. Robert Franklin Mehl (head of the Department of Metallurgical Engineering, Carnegie Institute of Technology, Pittsburgh) in recognition of his outstanding contributions to the science of metals and to metallurgical education; *Rosenhain Medal* for 1960, to Prof. Norman James Petch (William Cochrane professor of metallurgy, King's College, Newcastle on Tyne) in recognition of his outstanding contributions in the field of physical metallurgy; *W. H. A. Robertson Medal and Premium*, for 1959, to Dr. Maurice Cook and Mr. E. Swainson (formerly chairman and assistant manager, Technical Department, respectively, Imperial Chemical Industries, Ltd., Metals Division, Witton, Birmingham) for a paper on "Arc Melting of Reactive and Refractory Metals", published in the *Journal of the Institute of Metals*, 87, 161 (1958–59).

Wolfson Research Professorship of the Royal Society

THE Council of the Royal Society has accepted with thanks the offer of the Isaac Wolfson Foundation to create a special fund of £200,000 for the endowment of a research professorship, to be known as the Wolfson Research Professorship of the Royal Society. The chair is to be held by a distinguished British scientist, who will devote the whole of his time to research in such fields as the President and Council of the Royal Society may from time to time think appropriate. The first holder, who has yet to be appointed, will work in the physical sciences or their borderlines with biology. The financial provision allows for adequate scientific and technical assistance for the Wolfson research professor.

Herald Reactor at Aldermaston

THE research reactor *Herald* of the United Kingdom Atomic Energy Authority started operation on March 7 at the Atomic Weapons Research Establishment, Aldermaston. It will be used for nuclear research work requiring intense neutron beams and for irradiation-damage studies on reactor materials. *Herald* is a light water-moderated and -cooled tank-type thermal reactor designed to operate continuously at 5 MW. The core of the reactor is in the form of thin plates of enriched uranium–aluminium alloy suitably spaced in water. A high flux density of thermal and fast neutrons of the order of 10^{14} neutrons per $cm.^2$ will be achieved. The reactor was built for the Atomic Energy Authority by the Associated Electrical Industries/John Thompson Nuclear Energy Co., Ltd., and is an adaptation of the *Merlin* reactor.

U.S. Council for the Advancement of Science Writing

A U.S. Council for the Advancement of Science Writing has been established. Its purpose is to stimulate an increase in the quantity and quality of science news that is carried in all media of mass communications—newspapers, television, radio, magazines and books for the general public. It plans to do this by fostering research and training in science journalism, and by sponsoring discussions and seminars on science writing throughout the United States. The aim is to heighten the public's understanding and appreciation of the scientific enterprise. The Council, a non-profit corporation registered in the State of New York, was set up through the efforts of the National Association of Science Writers. Among those elected to the Council at its first meeting in New York City were: *President*, Earl

Ubell, science editor, *New York Herald Tribune*, who also is vice-president of the National Association of Science Writers; *Vice-president*, W. Bradford Wiley, president of John Wiley and Sons; and *Secretary Treasurer*, Prof. Hillier Krieghbaum, Harvard University. Further information can be obtained from the Department of Journalism, New York University.

Scales of Temperature in the Meteorological Office

THE Director-General of the Meteorological Office has decided that from January 1, 1961, the celsius (centigrade) scale of temperature will be used for all work carried out within the Office. At present, the Fahrenheit scale is used for surface observations (air temperature, dewpoint, soil and Earth temperatures); the change to the celsius scale for aerological work having been made some time ago. The decision to employ only the celsius scale in professional work is in line with the recommendations of the World Meteorological Organization, and its adoption not only brings the Meteorological Office into line with all other European services but also will greatly facilitate the international exchange of meteorological information. The change will affect only the internal practice of the Meteorological Office, and at present there is no intention to remove the degree Fahrenheit from the published forecasts, or from replies to inquiries.

Sudan Natural History Museum, Khartoum

THE report for 1956–58 of the Natural History Museum, Khartoum, is the first to be published since the control of this institution was assumed by the University. Since its formation in the 1920's the Museum has had a chequered career, and it was only recently that it passed to the University of Khartoum. This association has enabled the Museum to draw freely on the specialized knowledge of the University staff and to develop according to a deliberate long-term policy. On the other hand, the Biological Department of the University benefits considerably by easier access to the technical skill and knowledge possessed by the staff of the Museum. It also enables the staff of the University to use freely the collections and records in the Museum for teaching and research. The constitution of the Board for the Natural History Museum still awaits the adoption of a statute by the University Council; but this formality has not prevented full co-operation for some time past.

Communicable Diseases in School

COMMUNICABLE diseases are one of the commonest causes of school absenteeism. A survey by the World Health Organization examines the statutory regulations in force in fifteen countries (Geneva. 3*s*. 6*d*.). The measures prescribed, of which the exclusion from school of sick children and of contacts is paramount, are compared with the recommendations found in medical literature. Health authorities tend at present to substitute surveillance for stringent measures. Surveillance presupposes well-organized school health services and the existence of bacteriological laboratories. The subject-matter of the study published in the report is arranged under the following headings: legislation, exclusion measures, obligations of parents, teaching staff and health services; measures relating to school staff; and closure of schools. An appendix contains an analysis in tabular form, by country and by disease, of the statutory measures taken in countries the legislation of which is covered by the study. It is surprising to find that there is virtually no uniformity whatever in the measures prescribed for dealing with the occurrence in schools of diseases the epidemiology, evolution and method of spread of which are well known.

Caroline Haslett Exhibition for a Woman Engineer

THE Caroline Haslett Memorial Trust announces that the first award to be made by the General Section of the Trust will be an exhibition to enable a woman engineer to visit the U.S.S.R. for the purpose of studying the training, employment and prospects of Soviet women engineers. Applications for the exhibition are invited from women engineers in the United Kingdom whose training and experience would fit them to make the proposed study. The visit will be a short one and will probably take place in the early autumn of this year. After her return to Great Britain the exhibitioner will be required to prepare a report for the Trust and to lecture on her findings. The Caroline Haslett Memorial Trust was established in 1958 to commemorate the work of the late Dame Caroline Haslett. Application forms, which should be returned before March 31, can be obtained from the Secretary, Caroline Haslett Memorial Trust, 25 Foubert's Place, London, W.1.

Horticultural Co-operative Marketing Societies

THE Minister of Agriculture, Fisheries and Food, the Rt. Hon. John Hare, is setting up a departmental Working Party "To consider and report on the methods and forms of association between horticultural co-operative marketing societies and their members, and whether these might be modified so as to give the societies more stability and continuity in their supplies of produce". Mr. J. B. Godber, joint parliamentary secretary to the Ministry, will be chairman of the Working Party, and it is expected to have its first meeting in mid-March. It will seek evidence from the agricultural co-operative associations and their constituent societies on the horticultural side, and from other persons with a direct knowledge of the subject.

University News: Bristol

THE following are announced: *Appointments*, to become lecturer in aeronautical engineering, Mr. J. W. Flower; to become senior lecturer in physics, Dr. W. M. Gibson. *Grants*, from the Department of Scientific and Industrial Research, £15,000 for an electron microscope, and £3,850 for an investigation in the Department of Physical and Inorganic Chemistry on the electronic spectra of ionized hydrocarbons; from the Royal Society, £2,291 to the Department of Physical and Inorganic Chemistry for the purchase of a high-temperature resistance vacuum furnace in connexion with investigations on the synthesis and chemical properties of carbides, silicides, borides and phosphides of metals. Biological material to the value of £2,800 has been received by the Department of Pharmacology from the Bingham Oceanographic Laboratories, Yale University.

Cambridge

THE Council of St. John's College, Cambridge, will make a Kenneth Craik Research Award for the assistance of persons engaged in postgraduate research, preferably in physiological psychology. The value of the Award will be £450 a year. Applications should be sent to the Master, St. John's College, Cambridge, to reach him not later than April 4.

London

MR. P. T. BAUER, Smuts reader in Commonwealth studies in the University of Cambridge, has been appointed to the University chair of economics (with special reference to under-developed countries and economic development) at the London School of Economics and Political Science. Mr. C. B. Winsten, lecturer in the Department of Mathematics at the Imperial College of Science and Technology, has been appointed to the University readership in statistics tenable at that College.

Dr. J. H. Green, senior lecturer in physiology in Middlesex Hospital Medical School, has been appointed to the University readership in physiology tenable at that School.

Nottingham

DR. R. W. HEPBURN, lecturer in moral philosophy in the University of Aberdeen, has been appointed to the chair of philosophy, in succession to Prof. W. J. H. Sprott, who has been appointed to a newly established chair of psychology in the University.

University College of North Staffordshire

THE following have been appointed to senior lectureships as from October 1: Dr. D. E. Davies (physics), Dr. J. P. de C. Day (philosophy), Dr. D. S. Lees (economics), Dr. R. McWeeny (theoretical chemistry), A. C. Montefiore (political and moral philosophy), and W. M. Williams (geography).

Sheffield

IT is announced that Dr. H. Ruben has been appointed as senior lecturer in charge of the Department of Statistics. The following lectureships are also announced: Dr. D. M. Burley (applied mathematics); Dr. Margaret M. Platts (medicine); H. A. Salmon (physiology)

Announcements

SIR ALEXANDER FLECK, recently chairman of Imperial Chemical Industries, Ltd., has been elected president of the Society of Chemical Industry for 1960/61 in succession to E. J. Solvay, and will be installed at the annual meeting of the Society at Bristol in July.

THE Philosophical Society of Washington has announced the election of the following officers for 1960: *President*, L. R. Maxwell; *Vice-presidents*, L. M. McKenzie and R. D. Myers; *Corresponding Secretary*, F. N. Frenkiel; *Recording Secretary*, M. M. Shapiro; and *Treasurer*, L. Slack. Further information concerning the society may be obtained from F. N. Frenkiel, Applied Mathematics Laboratory, David Taylor Model Basin, Washington, 7, D.C.

IN written answers to questions in the House of Commons about day-release students in technical education, the Secretary of State for Scotland, Mr. J. Mackay, on February 23 gave the number of boys and girls under eighteen released by their employers in Scotland for part-time education in the year ended July 31, 1958, as 17,934 out of 163,000 estimated employed at the end of May 1958. For England and Wales in 1957–58, Sir David Eccles on February 24 gave the figures 191,889 boys out of 578,000 and 50,636 girls out of 585,000.

IN a written reply to a question in the House of Commons on February 24 regarding the professional staff employed by the Atomic Energy Authority, the Minister of Education, Sir David Eccles, as representing the Minister for Science, said that of 4,223 qualified staff, 32 were biologists, 863 chemists, 2 geologists, 239 mathematicians, 990 physicists, 275 metallurgists, 113 chemical engineers, 184 civil and structural engineers, 505 electrical engineers, 1 mining engineer, 1,014 mechanical and other engineers, and 5 held the B.Sc.(Tech).

A SYMPOSIUM on the Formation and Breakdown of Hæmoglobin will be held by the Association of Clinical Biochemists in conjunction with the Biochemical Society on March 26, at the School of Chemistry, University of Leeds. Further information can be obtained from Dr. J. H. Wilkinson, Westminster Medical School, Horseferry Road, London, S.W.1.

THE Society of Instrument Technology is arranging a series of one-day conferences. The first is to be held at the Grand Hotel, Bristol, on March 22, and is entitled "The Value of Instruments and Automatic Control to the Smaller Firm". Further information can be obtained from Mr. A. E. Hiden, 13 Chestnut Road, Long Ashton, Bristol.

NORWOOD Technical College announces that a short course on an introduction to Micro- and Semi-microchemical Methods will commence on April 1. The course will consist of ten weekly lectures and appropriate practical work designed to survey the branches of chemistry in which micro techniques are applied. Further information can be obtained from the Secretary, Norwood Technical College, Knight's Hill, London, S.E.27.

A MEETING of the British Food Manufacturing Industries Research Association is to be held in Edinburgh at the McEwan Hall, University of Edinburgh, on March 30. The theme of the meeting will be Science and Food, and besides various speakers an exhibition tracing the history of the canning of foods will be organized. Further information can be obtained from J. B. Pollard, British Food Manufacturing Industries Research Association, Randalls Road, Leatherhead, Surrey.

UNDER the auspices of the Shell International Petroleum Co., an annual course for science teachers will be held at University College, London, during April 5–8. The opening talk will be given by Lord James of Rusholme, high master of Manchester Grammar School. Three exhibitions concerning oil and related subjects will also be mounted. Further information can be obtained from Mr. J. A. G. Croxson, Shell International Petroleum Co., 1 Kingsway, London, W.C.2.

A THIRD symposium on aspects of agricultural meteorology will be held at Aberystwyth, on March 16, when the subject will be "Upland Climate and Land Usage, with Special Reference to Highland Britain". Memoranda prepared in connexion with the two earlier symposia, on "The Growing Season" and "Shelter Problems", are being reprinted. Further information can be obtained from Mr. James A. Taylor, Department of Geography and Anthropology, University College, Aberystwyth.

PLANNING IN SCIENCE

IN recent years there has been a great deal of discussion in many countries on whether it is possible to plan science, the effect of such planning on science and whether planning does not of necessity threaten the freedom of scientific research or limit the initiative of scientists. These issues were discussed at an international symposium sponsored by the Czecho-Slovak Union of Scientific Workers and held in Prague's Hotel International during September 19–22. Some hundred scientists were present—forty from abroad, from Great Britain, the Soviet Union, France, Brazil, India, Japan, Greece, Poland, Bulgaria, the German Democratic Republic, Rumania, Holland, Denmark, Portugal and North Korea. An observer from Unesco was also present.

The first and fundamental question raised was whether science can, by its nature, be planned at all. The participants in the symposium, without exception, agreed that planning the main tendencies of scientific research and ensuring that they are in harmony with the needs of the country and the development of science is not only possible to-day but also imperative. Naturally, the conditions for the planning of science are very different in a socialist society from those in a capitalist society. However, as pointed out by I. Malek (Czechoslovakia): "Even though scientific development in the capitalist countries has taken its course to date without purposeful and uniform planning, the majority of scientists and public figures responsible for the development of science do not doubt the necessity of planning in some form". P. L. Kapitza (U.S.S.R.) attributed recent spectacular Soviet scientific successes to planning: "The socialist State has great advantages in so far as the planned organization of science is concerned; this is demonstrated by the fact that it was the U.S.S.R. which had the first satellite and also was the first to reach the Moon. We do not think that the quality of our scientists is better than that of scientists of other countries. We are all people and we all know the same, think the same, have the same brains in our heads and our heads are of the same dimensions. But we were able to mobilize the means and the scientists and concentrate them on an important task".

The symposium then went on to discuss whether the planning of science may not become a serious obstacle barring creative research activity. Many of those present admitted that misgivings along these lines could indeed be justified in some circumstances. Care must be taken in formulating the plan, never to forget the individuals who were to carry it out. As M. M. Shemyakin (U.S.S.R.) put it: "Every bit of planning, particularly co-ordination and organization of complex research, must bear in mind the individual creative ability of the worker. Pure administration would not be in place".

Dealing with the potentialities and limitations in the planning of science it was pointed out that in scientific planning the occurrence of unexpected factors, particularly the possibility of basic discoveries in the natural sciences, must be reckoned with. As J. D. Bernal (Great Britain) pointed out: "The difficulties are very largely unforeseeable at the start and it is necessary to change course, to make use of any accidental advantage that crops up, even to abandon the whole attempt and to begin in some other direction. Even then you may not succeed in what you originally aimed at, though maybe instead you will have picked up something unforeseen and even more interesting and useful".

The problem of who should plan and guide scientific research was the subject of lively discussion during which the specific nature of planning in science, requiring special procedure and the widest participation of scientific workers, was stressed. P. L. Kapitza directed attention to the new type of scientific director needed to carry through large scientific projects. Such a director, whose role is creative rather than administrative, may be a very great man even though he himself does not work creatively in science. Interesting light on how scientific administrators are chosen in the U.S.S.R. was given by M. M. Shemyakin: "All heads of laboratories and institutes, all heads of departments at universities are elected every five years by competition in secret voting of the scientific councils. These scientific councils are set up from leading scientists of the given institute and other institutes. The directors of the Academy institutes are also elected for a period of five years from among the most important scientists, also by secret ballot of academicians and corresponding members of the Academy of Sciences of the U.S.S.R. In this way a truly democratic system of directing science is achieved and pre-conditions for control are created".

A great deal of the time of the symposium was devoted to the discussion of the difficult problem of how the results of scientific research can be appraised. It was generally agreed that the appraisal must be done by scientists and certainly not based on financial criteria. But scientists themselves are by no means able always to judge correctly whether a given line of research is likely to lead ultimately to promising results. In the final analysis, J. D. Bernal pointed out, the results of scientific research must be tested in practice. But sometimes, owing to insufficient farsighted appraisal, promising lines of research never get the possibility of practical test. This leads to a great waste of opportunity and a time-lag in putting valuable ideas into practice.

Some of the most interesting contributions to the symposium described how planning operated in practice in a number of countries. In the socialist countries the tendency to proceed from short-term and detailed plans to long-term planning of the main trends and tasks was general. In the U.S.S.R., after a very broad discussion, thirty problems of the natural and social sciences were designated which were recognized as the most timely for the years 1959–65. The planning of science is especially thorough in Czechoslovakia, where a national plan of research including all branches of science and embracing basic and applied research right up to the production stage has been formulated by the Czechoslovak Academy of Sciences. This body is also responsible for securing the financial and material means necessary for carrying it out. Plans for scientific research in Poland, Bulgaria, the German Democratic Republic and North Korea were described.

The only paper describing the organization of science in a Western country was given by E. H. S. Burhop (Great Britain), who pointed out that although an aggregate plan of scientific research in any sense comparable with the plans of the socialist countries does not exist in Great Britain, nevertheless, laboratories working in related fields co-ordinate their work and in the field of basic research this method of free co-operation appears to work satisfactorily. Probably applied research could benefit from more co-ordinated planning, but this met with some difficulty in a social system based on competitive private enterprise.

It would have added to the interest of the discussion if accounts had been available of the organization of science in other Western countries. Nevertheless, the conference was of great value to participants in that it revealed both the similarities and the contrasts between the problems facing scientists in countries having different social systems.

Ivan Malek

RECENT GEOPHYSICAL WORK IN BRITAIN

At its meeting on January 27, the Geological Society of London held a symposium on "Geological Implications of Recent Geophysical Work in the British Isles", under the chairmanship of the president, Dr. C. J. Stubblefield. The afternoon session, devoted to the study of igneous rock masses, was opened by Dr. M. H. P. Bott, who outlined the results of recent work on the variation of gravity over granitic intrusions[1-3]. Particular attention was paid to the form of the gravity profiles over two bodies with contrasting features, the Hercynian Bodmin Moor granite and the Caledonian Criffell granodiorite. In the former case a negative anomaly of about 50 milligals could only be interpreted in terms of a large density contrast (thought to be about 0·16 gm./cm.^{3}) between granite and country rock. In contrast, a relatively small negative anomaly of some 20 milligals was recorded over the Criffell granodiorite; the gravity profile could not be straightforwardly interpreted in terms of a uniform density contrast and it was necessary to postulate a major gradational increase of density within the igneous mass towards its south-eastern margin. These conclusions, as well as the space forms deduced for the two intrusions, were not out of harmony with the results of earlier field and petrological studies. It was inferred that both intrusions extended to depths of the order of 11–12 km., more than one third of the normal crustal thickness in continental regions.

Sir Edward Bailey questioned the assumption that there was no significant increase in the density of granites with depth, and quoted examples such as the Ben Nevis complex where the reverse had proved to be the case. Dr. A. H. Cook thought that since it was impossible to estimate densities at depth with accuracy, there was no valid evidence for denying the possibility of granite masses extending down to the Mohorovičić discontinuity. Prof. T. Murphy commented on areas of positive anomaly which flanked the gravity lows over certain Irish granites and stressed the need to consider the regional picture as a whole.

Dr. Bott also discussed the relationship which often appears to exist between the mass deficiency over a granite, calculated from the gravity anomaly, and the excess mass of the topography. This comparison led him to the suggestion that the high topography frequently associated with outcropping granites may sometimes be isostatically compensated, but within the crust rather than at its base.

Dr. D. J. Blundell described some recent investigations carried out to examine the extent to which the magnetic properties of igneous rocks might be of use in geological interpretation. He instanced his own study of the Lundy dyke swarm[4], in the course of which he was able to show that the mean direction of magnetization of the intrusions agreed with that of Tertiary lava flows. Another example cited was the work of Dr. D. Armstrong on some minor intrusives of Ayrshire[5]. This showed that the kylites and crinanites formed two separate suites with distinct polarization directions, the former corresponding to the known Permian direction, the latter consistent with a Tertiary age.

Palæomagnetic work on the Newer Basic Intrusions of Aberdeenshire gave rise to some discussion. Dr. Blundell and Prof. H. H. Read[6] interpreted the results as indicating that the Huntley, Insch and Belhelvie gabbros had been intruded in approximately their present attitudes and had not suffered subsequent deformation, a conclusion carrying important structural and petrogenetic implications. Prof. F. H. Stewart and Prof. R. M. Shackleton found it difficult to accept the palæomagnetic evidence as conclusive, pointing out the need for more detailed study of the geology of the Insch mass and of the effect of such features as the ore mineralogy and the thermal history on the magnetic behaviour of the rocks.

Dr. D. W. Powell quoted difference in magnetic dip between two Scottish Lower Carboniferous basalt lava sequences, both of which contained reversals, indicating polar movement during the period. He commented also on the effect of compression on rock magnetization as shown by laboratory experiments.

Mr. S. H. U. Bowie described the results of an airborne radiometric survey of Devon and Cornwall carried out for the Atomic Energy Division of the Geological Survey[7]. This had disclosed a relatively even level of radioactivity over the different granite masses, though individual members such as the Castle an Dinas phase of the Land's End granite, as well as kaolinized areas in general, were markedly low in activity. Acid igneous rocks gave much higher values than basic varieties and there was a general tendency for radioactivity to rise in the late differentiates of granites. On Lundy Island, however, Dr. A. T. J. Dollar had found the earlier granite to be the most highly radioactive member of the complex.

The evening session dealt with the study of sedimentary basins and their sub-surface geology. The results of seismic work in the Carboniferous basin of the East Midlands and the Hampshire Mesozoic and Tertiary basin were outlined by Mr. R. G. W. Brunston and Mr. N. G. O'Halloran. With increasing knowledge of the general structural picture and the need for detailed exploration and study of individual structures in the course of the search for oil within these basins, gravity and seismic refraction surveys were now being superseded by seismic reflexion work. In the East Midlands many pre-Permian folds had been discovered by this method and afterwards confirmed by drilling. A smaller number of pre-Albian folds had been found in Hampshire and Dorset and the margins of the basin more accurately delimited.

Dr. W. Bullerwell described the results of aeromagnetic surveys over central, eastern and southern England carried out under contract for the Geological Survey. Regions of positive anomaly were located in the northern parts of Leicestershire and Northamptonshire, in the area surrounding The Wash, along a magnetic 'ridge' extending roughly from Reading to Birmingham and in the English Channel south-east and south-west of the Isle of Wight. Comparison of the magnetic map with the gravity map of the same area, based upon data provided by the Geological Survey, University of Cambridge and the British Petroleum Co., revealed many regions in which gravitational and magnetic anomalies showed clear positional relationships, either directly or inversely correlated as regards amplitude. The general picture of the sub-Mesozoic surface of central England was suggestive of a block pattern in which the trends of such familiar structural lines as the Lickey, Nuneaton and Charnwood 'axes' were discernible.

The combined gravity and magnetic data, supported by the evidence of deep boreholes, afforded a suggestion of a sub-Mesozoic surface of acid igneous material at relatively shallow depth between Market Harborough and Bedford; and of a north-westerly trending geosyncline filled with Lower Palæozoic sediments beneath southern East Anglia, an interpretation in harmony with views previously expressed by Sir Edward Bullard and others after seismic investigations in this area[8].

Recent research, outlined by Prof. W. B. R. King and Prof. W. F. Whittard, supplemented previous knowledge of the geological history of the English Channel and Western Approaches[9–11]. In the central and eastern Channel, seismic work had proved high-velocity rocks at much less depth than anticipated. It appeared that the Paris Plage Palæozoic ridge was of greater extent than formerly thought and must virtually have separated the Hampshire and Paris basins. Probably the Trias was absent over this ridge and the Jurassic succession much thinner than that in the Portsdown and Henfield borings to the north and the Pays de Bray to the south. In the Western Approaches, Prof. Whittard inferred a series of tongue-shaped outcrops ranging from Upper Cretaceous to late Tertiary, with the younger formations appearing progressively towards the south-west. On the continental slope indurated blocks of chalky material containing Recent, or at earliest Pleistocene, Foraminifera had been dredged from a depth of 500 fm.

Among other contributions dealing with marine geophysics was one from Dr. R. F. King describing seismic, gravity and magnetic surveys in the northern part of Cardigan Bay, supplementing previous work by Dr. D. W. Powell[12]. The density and seismic velocity of the upper layers suggested the presence of clays of late Mesozoic or Tertiary age. Mr. G. Armstrong described the use of the sub-bottom depth recorder (or 'sparker'), essentially a broad-band, high-powered, echo-sounder, using as a sound source either an electric spark or a small explosion. With this instrument the National Coal Board had obtained a reasonably comprehensive picture of the fault pattern, disposition of strata and distribution of igneous dykes off the coast of Northumberland and in the Firth of Forth. An oblique echo-sounder system which recorded reflexions from the sea-bed from a low-angle beam had been successfully applied for geological purposes in Weymouth Bay by Dr. D. J. Donovan and Mr. A. H. Stride.

A feature of the symposium was the variety of the techniques discussed and the complementary nature of the geological evidence obtained by different methods. It was noteworthy that many of these (aeromagnetic, aeroradiometric, 'sparker', 'oblique asdic') employed continuous recording along a chosen profile. Increasing use of such methods is an important trend in applied geophysics at the present time and their advantages both in speed of operation and for interpretation were pointed out towards the close of the evening by Dr. Bullerwell.

J. H. Taylor

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[1] Bott, M. H. P., *Geol. Mag.*, 90, 257 (1953).

[2] Bott, M. H. P., *Quart. J. Geol. Soc.*, 112, 45 (1956).

[3] Bott, M. H. P., Day, A. A., and Masson-Smith, D., *Phil. Trans. Roy. Soc.*, A, 251, 161 (1958).

[4] Blundell, D. J., *Geol. Mag.*, 94, 187 (1957).

[5] Armstrong, D., *Nature*, 180, 1277 (1957).

[6] Blundell, D. J., and Read, H. H., *Proc. Geol. Assoc.*, 69, 191 (1959 for 1958).

[7] Bowie, S. H. U., Proc. 2nd Int. Conf. Peaceful Uses Atomic Energy, Geneva, 2, 787 (1958).

[8] Bullard, E. C., Gaskell, T. F., Harland, W. B., and Kerr-Grant, C., *Phil. Trans. Roy. Soc.*, A, 239, 29 (1940).

[9] Hill, M. N., and King, W. B. R., *Quart. J. Geol. Soc.*, 109, 1 (1953).

[10] King, W. B. R., *Quart. J. Geol. Soc.*, 110, 77 (1954).

[11] Day, A. A., Hill, M. N., Laughton, A. S., and Swallow, J. C., *Quart. J. Geol. Soc.*, 112, 15 (1956).

[12] Powell, D. W., *Quart. J. Geol. Soc.*, 111, 375 (1956 for 1955).

EVOLUTION OF ARBORVIRUS DISEASES

THE arborviruses (*ar*thropod-*bor*ne viruses) are those animal viruses which multiply in the tissues of the arthropod host and are transmitted in its saliva. The purposes of a symposium which had been arranged by the Royal Society of Tropical Medicine and Hygiene, and was held in London on January 21, were as follow: (1) to try to fit the arborviruses into current evolutionary theory; (2) to review current knowledge of arborviruses and suggest profitable lines of thought and investigation; (3) to interest workers in related disciplines, notably zoologists.

The first two papers dealt mainly with the origin and long-term evolution, the third mainly with evolution observable in our own time.

Prof. C. D. Darlington discussed the relationship between heredity and infection in plant, insect and animal viruses. Human viruses are particularly fruitful for study because while viruses are the most rapidly evolving of parasites, man is the most rapidly evolving, most varied and most widely distributed of higher organisms. Because viruses are wholly parasitic we have to consider the relations of harm or benefit between hosts and parasite. At the virus-level, the fundamental distinction in the basis of propagation is between the nucleic acids involved. In plants the two extreme forms of ribonucleic acid particle are the purely hereditary plasmagene of male fertility and the purely infectious particle of tobacco mosaic virus. Particles which are artificially transmissible by grafting, but not spread by natural infection, may be called proviruses, while viruses which apparently appear spontaneously or by chemical or physical treatment may be called new viruses. When vectors transplant viruses to new species the behaviour of the virus may change greatly, and when a vectored virus is artificially transmitted in series it may lose the capacity to infect the vector. A virus can become a harmless nucleoprotein (become latent) : or something hereditarily transmitted in the cytoplasm or generated by the cell in development may become infectious. Latent viruses which resemble the plasmagene in behaviour may arise from active viruses either in the capital host or the vector and may or may not be transmitted through the egg. Further evolution may lead to loss of infectiousness, as with Kappa in *Paramecium*, or the hereditary vector stage may be lost and only direct infections between capital hosts remain. Phages have evolved on a basis of alternating heredity and infection and it is possible that some viruses of higher organisms may also have done so. Heredity and infection seem, however, to be strict alternatives, for the viruses of higher organisms and hereditary particles are likely to persist only when they confer a benefit on the host. On the other hand, infectious particles owe their maintenance to the benefit they extract from the host, and harm to the host is relatively unimportant. The selection pressures are, therefore, opposed and when a hereditary particle becomes infectious they are switched.

Mr. P. F. Mattingly presented the view that the evolution of arborviruses has depended very largely on the ecology of their arthropod and vertebrate hosts. Despite their isolation in host tissues, each individual virus may be credited with an ecology of its own, just like other organisms. To appreciate this, secondary accretions such as adventitious transfer to man and occurrences in atypical hosts or vectors must be disregarded. When it was introduced into the New World, yellow fever rapidly found an ecological milieu very closely resembling that from which it had been liberated in Africa. The need seems to have been primarily for arboreal primate hosts. *Haemagogus* were available, in place of forest *Stegomyia*, and adaptation to these may have been relatively easy. Similarly the principal vectors of Wesselsbron virus in the low and high veldt (*Neomelaniconion* and *Ochlerotatus* respectively) are taxonomically distinct, but phenologically closely similar.

The present-day requirements for the establishment of a mosquito-borne virus appear to be the coexistence of warm-blooded vertebrates, angiospermous plants and blood-sucking mosquitoes. The two former are known to have evolved more or less together during the latter part of the Mesozoic and the viruses and their mosquito vectors probably evolved with them. The necessity of fitting into a complex ecological system may have slowed the rate of evolution compared with some other viruses. The fact that primate malaria can be transmitted only by anopheline mosquitoes while bird malaria is readily transmitted by culicines in the laboratory and, presumably, also in Nature, suggests that the anophelines originated as mammal feeders and the culicines, at least in part, as a bird-feeding group. The few known cases of virus transmission by anophelines, other than *Kerteszia*, appear to be adventitious or secondary. Basic maintenance cycles involve culicines only, except possibly in the subgenus *Kerteszia* which in some ways are more culicine than anopheline in their ecology. These facts suggest that the primitive mosquito-borne viruses were bird rather than mammal viruses. *Aëdes*-borne viruses are confined to the tropics. In the subtropics they are replaced by *Culex*-borne viruses and these in turn are replaced by tick-borne ones in the northern steppes. The explanation seems to be that *Aëdes* hibernate and æstivate in the egg, *Culex* as adults. In the relatively severe conditions prevailing in the subtropics hibernating adult mosquitoes appear to be necessary for the maintenance of virus. Farther north, only the resistant egg will serve for hibernation and *Aedes* become the dominant mosquitoes. But in the absence of transovarian transmission these are unsuited to the maintenance of virus, and mosquito-borne viruses are replaced by tick-borne ones. On the assumption that West Nile is the most primitive of the group *B* viruses, a picture can be drawn of invasion from subtropical swamp habitats to the northern steppes on one hand, and to a variety of tropical habitats on the other, with transfer to ticks and to *Aëdes* respectively. A simpler and more coherent picture would result from the assumption that the Russian spring–summer viruses were the most primitive, and the arguments previously adduced are worth reconsideration on this basis.

Dr. C. E. Gordon Smith concentrated mainly on the effects of intentional or unintentional human interference on the short-term evolution of arborviruses. About a third of the known arborviruses cause human disease ranging from severe encephalitis to mild fever—the remainder are still in search of a disease, but there are also many human febrile diseases which may be in search of a virus. Arborviruses are most numerous in the tropics and become progressively fewer as we go north or south to subtropics and temperate zones.

The viruses are usually maintained in their wild reservoirs without overt signs, and infections of man, and epidemics in man, spill over from these reservoirs under suitable conditions. Such infections are, however, without significance to the continued survival of the virus.

When a virus is in equilibrium with its hosts and environment (endemic or enzootic), the strain present is the one whose characteristics most perfectly fit the particular ecological complex. Under such conditions a virus mutant is very unlikely to be better adapted for survival than the parent form and is unlikely to succeed. Any change in the number or species of hosts or in the environment, however, provides conditions which may favour a new mutant, which may, by chance, be more or less virulent for man than the parent form. The sudden appearance of Kyasanur Forest disease in western India in 1955, and of a hæmorrhagic 'dengue' in the Philippines and Thailand in 1956, suggest that changes of these sorts must have occurred.

Among man's major activities the clearance of forest and introduction of new hosts to cleared areas are of outstanding importance. Unsuspected viruses may be liberated to establish themselves elsewhere. Russian spring–summer encephalitis became an important disease in Russia only when the taiga forests were cleared and settled.

Irrigation and subsequent agriculture in California brought changes in the flora and fauna which were responsible for epidemics of encephalitis, and increased agricultural activity in Tongaland appears to have led to greatly increased arborvirus activity there.

With the widespread use of insecticides, resistant arthropod strains emerge, and it is unlikely that they do not also differ from the parent form in other respects, such as vector efficiency for arborviruses. Arthropods do not appear to be adversely affected by arborvirus infection and it is not known whether their defence mechanisms interact with the viruses. In vertebrates the specific defence is antibody formation, but there are also the non-specific processes of inflammation and fever. Inflamed tissue is acid, thus a strain of virus with greater resistance to acid and which multiples better over 100° F. may prevail. In the arthropod, the length of the extrinsic incubation period is dependent upon temperature; thus, on the other hand, viruses which multiply better at, say, 70° F., might have an advantage.

A strain of virus capable of remaining latent in vertebrates for long periods might be maintained through periods when arthropods are not sufficiently numerous. Such latent infections with western equine virus have been demonstrated in birds and the stress of migration or ovulation might permit the virus to circulate when mosquito populations are becoming active in a temperate climate.

Opening the discussion, Dr. C. H. Andrewes suggested that there are two kinds of evolution to consider: the short-term with changes such as have been best seen in influenza, and the long-term, in which viruses as a whole may be very old and relatively stable. Viruses have a unique form of replication, which does not suggest that they are derived from host components. The ticks being probably more primitive than the mosquitoes might be more likely to have been the original hosts.

Several other speakers suggested pointers to the ticks as original hosts. Prof. D. S. Bertram pointed out that the one-layer covering of tick eggs may make them more susceptible to transovarial infection than mosquito eggs. Dr. M. G. R. Varma said that it appeared that while mosquito-borne viruses could multiply in ticks, tick viruses could not in mosquitoes. Dr. P. B. Stones suggested that if mosquitoes were the origin, then viruses ought to have been isolated from male mosquitoes: he reported an isolation of Semliki forest virus from a male *Eretmapodites* in Lagos.

Dr. J. S. Porterfield suggested that Semliki Forest virus may be the most primitive member of group *A* of arborviruses—a number of very closely related forms have now been isolated in Africa. He thought that Israel turkey virus may be a recent derivative of West Nile virus possibly resulting from man-made ecological changes.

On an analogy with transduction in bacteriophage, Dr. P. H. A. Sneath suggested that viruses are not true parasites, but may be partially derived from the reproductive mechanisms of their hosts.

C. E. GORDON SMITH

RED DEER AND VEGETATION IN NEW ZEALAND

AN expedition visited Lake Monk in the Cameron Mountains of Southern Fiordland, New Zealand, during March 26–April 5, 1957, to study the inter-relations of birds and mammals, particularly deer, with the vegetation (New Zealand Department of Scientific and Industrial Research, Bull. 135).

The vegetation was classed broadly into forest, scrubland, grassland and bogs. Mice and stoats are the only small animals in the area; the former are scarce and the latter are probably more widely distributed; from an examination of droppings they appear to be feeding mainly on birds. Red deer (*Cervus elaphus*) are thought to have reached the area about 1920, and to have built up a peak population between 1943 and 1950. They have since declined in numbers and are probably still declining. The population was composed of 50 stags to 35 fawns to 100 hinds, which are unusually low ratios; the deer were all in very poor condition.

The amount of grazing on the fern *Polysticum vestitum* was strongly correlated with the density of deer fæces, and could be used to provide an alternative index to deer abundance.

Counts of deer fæces showed that the animals in this area were spending more time in the forest than in the open grassland, although their preferred food at the time of year studied is grasses and forbs, but storms probably prevent the deer from spending as much time in the open as they would otherwise do. In the forest the beech seedlings are heavily grazed, but once established they continue to grow slowly. The saplings are hedged by deer; but the full amount of each year's growth is not removed and eventually a shoot grows above the browse-level There are sufficient pole-stage beech to replace the present generation of mature trees, and enough of the younger stage to ensure the continuance of the forest, although possibly with a lower density of trees.

The second-tier species in the forest, such as *Nothopanax colensoi*, *Griselinia littoralis*, and *Coprosma foetidissima*, have little resistance to browsing and the young stages of these species are only found in places inaccessible to deer. In the sub-alpine scrub there has been a widespread destruction of *Olearia colensoi*: grassland areas have been heavily used but show signs of recovery. On boggy areas the trampling of hooves has bared the ground around wallow holes.

The investigators concluded that red deer provide the dominant animal influence on the vegetation; but it is an influence superimposed on long-term trends due to climate and on short-term successional changes. The composition of the vegetation has been and is being altered by them in such a way that there is a marked increase in unpalatable plants, and a corresponding decrease in palatable plants. The resulting environment is relatively unfavourable to deer. In spite of these changes in the composition of the vegetation the forest, as a protective forest, is in no danger of being eliminated by deer; the total vegetative cover is not decreasing, and soil erosion in the area is negligible.

In the light of these findings an attempt is being made to define and clarify the wild-life problems arising from the use of this area as a national park.

THE LONG ASHTON RESEARCH STATION

THE volume of work reported in Long Ashton's annual report* is now so large that some selection is necessary to keep a review within manageable proportions.

Growth of apple fruits is dependent on the presence of seeds, but D. L. Abbott finds that this is so only during seven weeks after petal fall. C. Bould worked on the nutritional aspects of fruit production and found that yield and growth responses of Royal Sovereign strawberries in relation to nitrogen, phosphate and potash were those to be expected from leaf analyses of non-manured plants at the fruiting stage. This should provide a useful practical approach to the vital question of soil nutrient needs. Pollination is a necessary factor in fruit production, and it is surprising that relatively little is known about the distance over which a pollinator variety is effective. R. R. Williams found it to be about 90 ft. in one orchard, but only 50 ft. in another. A survey by A. I. Campbell and R. R. Williams of eighty orchards in Hereford and Somerset shows that difficulties encountered in headworking cider apple trees are in part due to apple virus diseases. It is sometimes necessary to clear old tree-stumps from land; E. W. Hobbis and J. S. Coles have investigated various possible rotting agents—aqua regia, caustic soda, bleaching powder, and a nitrogen–phosphate fertilizer. Only the last material showed any effect after four years. Workers in the pomology section also discuss the technique of measuring the assimilation of radioactive carbon by leaves, a survey and descriptions of perry pear varieties, and the forecasting of harvest date in blackcurrant varieties from their flowering dates.

A series of papers on spray application problems gives methods for the determination of spray residues —of gamma-BHC (J. A. Pickard), of microgram quantities of dieldrin (E. J. Skerrett and E. A. Baker), and of rotenone in *Lonchocarpus* (D. V. Richmond). A mercury content of 1 p.p.m. was found in coffee berries on trees which had received thirteen applications of mercury fungicide (J. A. Pickard and J. T. Martin). The plant cuticle is the immediate receptor for spray fluids, and J. T. Martin reviews current work on its nature and composition in several fruits and vegetable crops. Uptake of a fungicide (DMTD) by fungal spores (E. Somers), and the metabolism of certain aromatic compounds by fungi (D. Woodcock and R. J. W. Byrde) are also discussed. There has been quite a revolution in outlook on methods for spray application, and T. E. Cobbald briefly describes a prototype low-pressure twin-fluid atomizer. Fluorescent materials as tracers in spray liquids, and the vibrating jet method for the determination of dynamic surface tensions are two methods of assessing spray performance discussed in the report. Considerations of actual disease are mainly concerned with *Gloesporium* fruit rot, and entomology is represented by a paper on seasonal distribution of four species of leaf hoppers on apples. An interesting result of importance overseas is the finding of the fungus *Trachysphaera fructigena* on Jamaican bananas. The infection appears to have occurred in ripening rooms, from fruit from another source.

There are papers on cider and fruit juices and on domestic fruit preservation, as in previous years, and a list of 46 publications during 1958. It is announced in the introduction that the Agricultural Research Council's Unit of Plant Nutrition at Long Ashton was disbanded in September 1959. Members of its staff have, however, been absorbed into the work of the Station.

JOHN GRAINGER

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* Annual Report of the Agricultural and Horticultural Research Station (The National Fruit and Cider Institute), Long Ashton, Bristol, 1958. Pp. 190+18 plates. (From the Station, 1959.) 15s. post paid.

INFLUENCE OF NITROGEN-CONTAINING BASES ON THE RATE OF THE ELECTRODE PROCESSES AT THE DROPPING MERCURY ELECTRODE

By MRS. V. VOLKOVÁ

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A SPECIFIC effect of gelatin, amines, *p*-aminobenzoic acid esters of amino-alcohols and nitrogen-containing heterocyclic compounds such as various pyridine derivatives, alkaloids, etc., has been found[1,2] during the polarographic reduction of many organic compounds.

In respect of the polarographic waves, the effect on the reduction of substances capable of dissociation was of two kinds: on one hand, an influence on the height of the first wave, on the other hand, a considerable shift of the second wave to more positive potentials, often by several hundreds of millivolts.

The first case was observed chiefly with very weak acids, containing a heterocyclic nitrogen atom such as pyridine carboxylic acids, quinaldinic acid and oxyquinolines, and with corresponding aldoximes, further with α-Br-carboxylic acids, phthalic acid and benzaldehyde, the shift of the second wave being found with many reducible weak acids to such an extent that only a single wave was visible.

The effect on the first wave was originally explained by a reaction controlled by the recombination of the anion with the nitrogen-containing cation as proton-donor[1]. The recombination-rate constants thus obtained were of the order 10^{8}–10^{10} except with atropine and some other alkaloids, where considerably larger values were obtained which depended on the concentration of the nitrogen compound.

Since the above-mentioned catalysts are substances of strongly adsorptive character, attention has been directed to the study of the role that this property takes in the problem studied. The conception of the

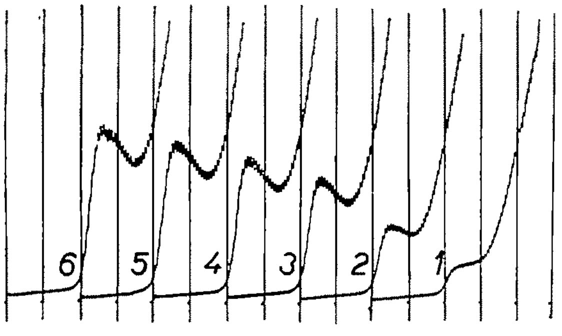

Fig. 1. Effect of amino-pyridines on the first wave of 10^{-4} *M* isonicotinic acid; borate buffer *p*H 10; concentration of 2-amino-pyridine: (1) 0, (2) 0·5 × 10^{-2} *M*, (3) 1·5 × 10^{-2} *M*, (4) 2 × 10^{-2} *M*, (5) 2·5 × 10^{-2} *M*, (6) 3 × 10^{-2} *M*; each curve starts at − 1·0 V. (vs. S.C.E., 0·2 V./absc.)

adsorption of the catalyst was supported in this case by the fact that the catalytic influence tended towards a limit with respect to the concentration of the catalyst. The influence of the adsorption of cations has been observed before in the polarographic reduction of inorganic anions; recently this case was thoroughly discussed by Frumkin[3]. Evidence for the adsorption occurring in our case as an accumulation of the catalyst at the electrode surface has been obtained from the study of *i–t* curves. When alkaloids, which have a large adsorption coefficient, were used as catalysts the effect of acceleration of the electrode process with time (a distinct inflexion on *i–t* curves) occurred at potentials where the mean current represented less than a quarter of the limiting current, but in the presence of pyridine derivatives and gelatin the *i–t* curves exhibited always the monotonous kinetic course.

The shape of *i–t* curves for different concentrations of the alkaloid (see Fig. 2 with phenylglyoxylic acid in the presence of atropine) corresponds to the concept of acceleration of the electrode process by the adsorption of a polarographically inactive substance with a large adsorption coefficient on the electrode surface. Because of the large adsorption coefficient, a certain period of time has to pass before the quantity of surface-active substance necessary for the saturation of the electrode surface is transferred to the electrode by diffusion.

Due to this increase of the surface concentration of the catalyst at the electrode the rate of the electrode process is also gradually increased until the limit concentration on the electrode is reached. The influence of the adsorption of an electrode-inactive substance with a large adsorption coefficient on the electrode processes of depolarizers has been theoretically solved by Weber, Koutecký and Koryta[4,5]. For the instantaneous current i at the beginning of the rising portion of the polarographic wave for periods of time smaller than ϑ (the time when the surface coverage has been attained) and for cases in which the rate constant at the occupied surface k_1 is distinctly larger than that at the free surface k_0 the following relation, $i \sim t^{7/6}$, holds evidently only in the case when the effective rate constant k_{ef} is a linear function of θ, the degree of the coverage of the electrode, and k_1 does not change with this degree of the coverage of the electrode. The relations between these quantities are as follows[6]:

$$\theta = \frac{\Gamma}{\Gamma_k} = \left(\frac{t}{\vartheta}\right)^{1/2}; \quad \vartheta = 1{\cdot}85 \times 10^{6} \times \Gamma_k^{2} \times c_A^{-2} D_A^{-1}$$

where Γ and Γ_k are the instantaneous and maximal concentrations of the catalyst adsorbed at the electrode surface, c_A the concentration of the surface-active catalyst in the solution, and D_A its diffusion coefficient.

The *i–t* curves at the beginning of the rising portion of the wave of the anion of phenylglyoxylic acid in the presence of alkaloids represent the most suitable example for the relations mentioned above. Although the exponent for $t < \vartheta$ was larger than 7/6 (up to 1·6) the dependence of ϑ on c_A^{-2} was found to be linear. Putting $D_A = 6{\cdot}4 \times 10^{-6}$ cm.²/sec. the area occupied by one molecule of atropine has been calculated as 93 Å.². The higher value of the exponent can be explained by the assumption that the adsorption is a kinetic process, or by the change of k_1 during the drop time with the change of θ or by supposing that for other reasons (the change in the value of the potential ψ_1) k_{ef} is not a linear function of θ. This will be the object of further investigations.

In the case of substances with a small adsorption coefficient the adsorption equilibrium is strongly shifted in favour of the non-absorbed form, so that Γ remains constant during the drop time and if the equilibrium is established very rapidly Γ can be expressed by any adsorption isotherm. Such a case can be solved for mean currents as in a normal slow electrode reaction[7]. If k_{ef} is a linear function of θ for suitably small concentrations for which θ is

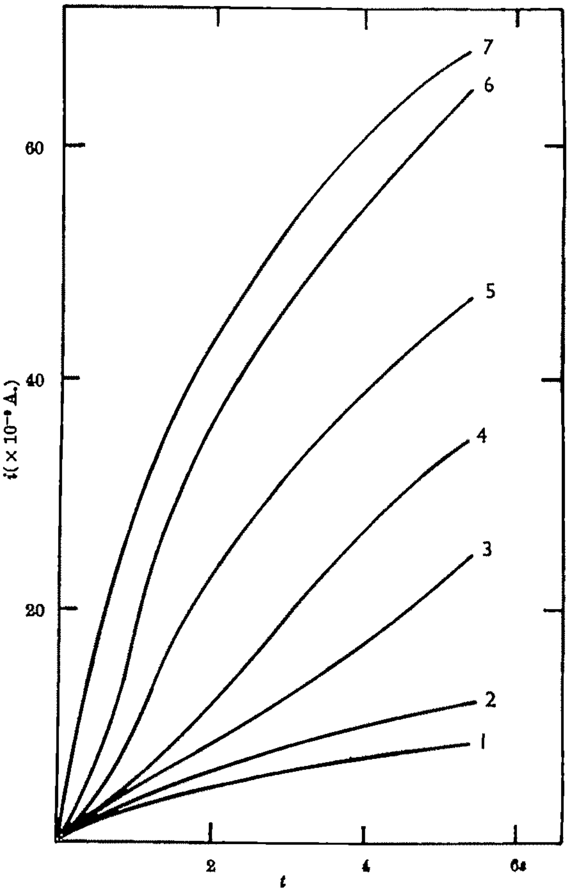

Fig. 2. Effect of alkaloids on *i–t* curves of 4 × 10^{-3} *M* phenylglyoxylic acid (as depolariser); borate buffer *p*H 8·6. *E* = − 1·04 V. (vs. S.C.E.); concentrations of atropine: (1) 0, (2) 0·4 × 10^{-6} *M*, (3) 3·3 × 10^{-6} *M*, (4) 5 × 10^{-6} *M*, (5) 8·3 × 10^{-6} *M*, (6) 13·3 × 10^{-6} *M*, (7) 23·3 × 10^{-6} *M*

also a linear function of the concentration of the catalyst in the solution and at a constant pH, then: $\frac{i}{i_d - i} \sim C_{\text{kat}}$. A good agreement of experiments with this relation has been found for some derivatives of pyridine.

A plausible explanation of the acceleration of the electrode process is as follows: since in all investigated cases the adsorption occurred at potentials at which the electrode has a negative charge, it can be assumed that the surface-active substances are adsorbed chiefly as cations. The circumstance that a very distinct acceleration of the electrode process occurs if anions are reduced or undissociated substances with the possibility of forming zwitterions such as *iso*nicotinic acid, hydroxyquinolines, pyridinealdoximes, etc., would support the assumption that a closer approach of the depolarizer to the electrode is made possible, perhaps because of a tendency to form an adduct. The distinct increase of the current of the second wave of phthalic acid which is ascribed to the reduction of the undissociated acid[8] could be explained by the assumption that the acid exists in the form with separated charges the electrode reaction of which is catalysed. The idea of thus influencing the reduction is supported also by the finding that in the reduction of a particle with a positive charge such as in the reduction of the cation of phthalic acid the presence of a surface-active substance as a cation causes a lowering of the current. There is no tendency towards the formation of an adduct in this case; on the contrary, the presence of the cation at the electrode surface causes a repulsion of the depolarizer with a positive charge and therefore a retardation of the electrode process must take place.

The details will be published elsewhere.

[1] Volková, V., Czechoslov. Hungar. Polarograph. Conference, Prague (July 1958).

[2] Volková, V., Second International Polarographic Congress, Cambridge (August 1959).

[3] Frumkin, A. N., *Trans. Faraday Soc.*, **55**, 156 (1959).

[4] Weber, J., Koutecký, J., and Koryta, J., *Z. Elektrochemie*, **63**, 583 (1959).

[5] Weber, J., Second International Polarographic Congress, Cambridge (August 1959).

[6] Koryta, J., *Chem. Listy*, **47**, 340 (1953); *Coll. Czech. Chem. Comm.*, **18**, 206 (1953).

[7] Koutecký, J., *Chem. Listy*, **47**, 323 (1953); *Coll. Czech. Chem. Comm.*, **18**, 597 (1953).

[8] Ryvolová, A., and Hanuš, V., *Chem. Listy*, **50**, 46 (1956); *Coll. Czech. Chem. Comm.*, **21**, 853 (1956).

SOME SIMILARITIES AND DISSIMILARITIES IN THE STRUCTURE OF THE SKIN AMONG THE MEMBERS OF THE SUBORDERS ODONTOCETI AND MYSTACOCETI (CETACEA)

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THE members of two cetacean suborders, while possessing a number of similar features, show at the same time some essential differences. Obviously, the same relations must exist in the structure of their skin as well.

Up to now no attempts have been made to compare the structure of the skin in those suborders, although the cutaneous covering of some cetacean species has been studied by several investigators[1].

As material for the work reported here, samples of the skin from the following species of Cetacea were used: *Eubalaena glacialis* Bonaterre (obtained in 1955 by permission of the Minister of Fisheries *N*0113/2010, dated March 3, 1955), *Balaenoptera physalus* L., *B. borealis* Lesson, *B. acutorostrata* Lacépède, *Physeter catodon* L., *Berardius bairdii* Steineger, *Delphinus delphis* L., *Lagenorhinchus obliquidens* Gill and *Orcinus orca* L.

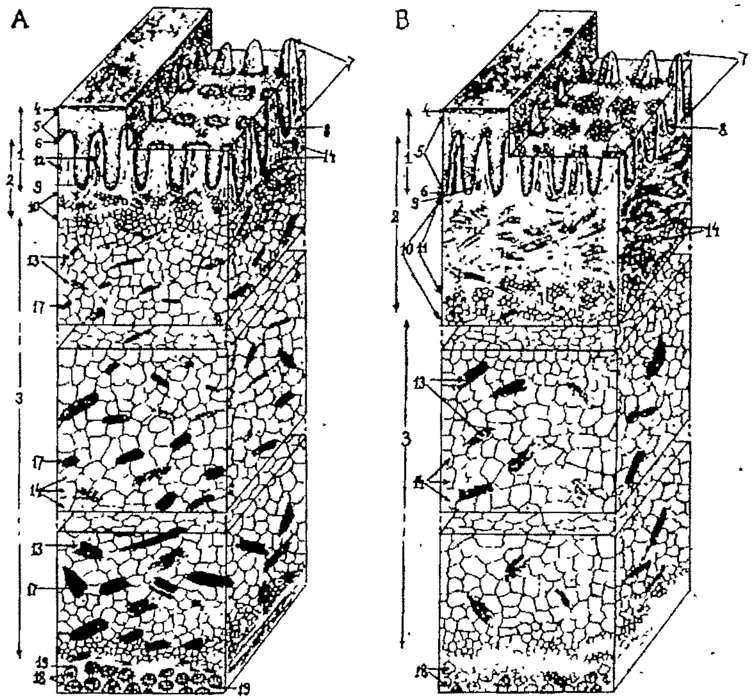
Most samples were collected during the expedition sent by the Institute of Oceanography of the Academy of Sciences of the U.S.S.R. (head of the expedition, S. Klumov).

Samples taken across the whole skin up to subcutaneous muscles were embedded in 'Celloidin'. Sections made by Christeller's method were stained with hæmotoxylin-eosin, after Van-Gieson and with resorcin-fuchsin.

Peculiarities of the cetacean skin common to all members of that order will be considered first. The cutaneous covering of Cetacea differs from that of other mammals by the absence of a continuous pelage and skin glands. Their skin consists of the following layers: the epidermis (Figs. *A*1, *B*1), the dermis (Figs. *A*2, *B*2) and the subcutaneous fat tissue (Figs. *A*3, *B*3). The limit between two latter layers is slightly marked. The epidermis, though reaching considerable thickness, consists of only three layers: the outer one, called conditionally the corneous layer (cornification does not take place in it and its cells retain nuclei) (Figs *A*4, *B*4); the underlying stratum spinosum or prickle cell layer (Figs. *A*5, *B*5); and the deepest basal layer (Figs. *A*6, *B*6). On the inner surface of the epidermis numerous alveoli are distributed, in which dermal papillæ rise vertically upwards (Figs. *A*7, *B*7). The epidermis may be pigmented, the pigment in the form of finest granules being disposed in the epidermal cells and localized above the cell nucleus. At the limit between the epidermis and dermis melanophores sometimes occur.

The dermis of Cetacea consists of dermal papillæ (Figs. *A*7, *B*7) and of a sub-papillary layer (Figs. *A*10, *B*10) in which a non-adipose layer (Fig. *B*11) adjoining the epidermis may occasionally be seen. The dermal papillæ are formed of thin bundles of collagenic fibres rising vertically upwards along the long axis of the papilla, and of a few elastic fibres running in the same direction. Small blood vessels rise to the top of the papilla (Fig. *A*12). No fat cells are present in the dermal papillæ. In the sub-papillary layer of the dermis bundles of collagenic fibres (Figs. *A*13, *B*13) are situated at various angles to the surface of the skin, forming a more or less dense network. Elastic fibres may be numerous. The thickness of the latter as well as that of the bundles of collagenic fibres is considerably greater than in papillæ. Fat cells (Figs. *A*14, *B*14) are disposed either immediately at the epidermis itself or at some distance from it, in which case a non-adipose layer (Fig. *B*11) with a very compact network of bundles of collagenic fibres will be formed. The dermis passes gradually into a subcutaneous fat tissue (Figs. *A*3, *B*3) in which collagenic fibre bundles (Fig. *A*13, *B*13) are disposed far from each other, the whole space between them being filled up with enormous accumulations of fat cells (Figs. *A*14, *B*14). With their removal from the epidermis the bundles of collagenic fibres become thicker and the fat cells grow larger, decreasing only in size near subcutaneous muscles. Elastic fibres and large blood vessels are but few in the subcutaneous tissue. A vast number of capillaries pass between the fat cells.

In describing differences of the skin in two suborders of the order Cetacea, two families, Balænopteridæ and Balænidæ, among Mystacoceti, have to be considered separately because the skin of the latter combines characters of both the Odontocete and Balænopteridæ. The skin of ***Eschrichtius gibbosus***, family Eschrichtiidæ, has not been studied.



A scheme of the skin structure in Mystacoceti (*A*) and Odontoceti (*B*)
1, Epidermis; 2, dermis; 3, subcutaneous fat tissue. Epidermal layers: 4, corneous; 5, stratum spinosum or prickle cell layer; 6, basal; 7, dermal papillæ; 8, longitudinal septa of epidermal alveoli; 9, transversal septa of epidermal alveoli; 10, subpapillary dermal layer; 11, non-adipose part of subpapillary dermal layer; 12, blood vessels in dermal papillæ; 13, bundles of collagenic fibres; 14, fat cells; 15, epidermal projections extending into the tissue of dermal papillæ; 16, oval form of the diameter of epidermal alveoli; 17, bundles of elastic fibres; 18, bundles of striated muscle fibres in subcutaneous muscles; 19, elastic fibres surrounding bundles of striated muscle fibres in subcutaneous muscles

On a tangential section of epidermis the epidermal alveoli of Balænopteridæ and of the surface-swimming Odontoceti have the form of an oval (Fig. *A*16), the long axis of which is directed along the animal's body. In deep-diving Odontoceti (*P. catodon* and *B. bairdii*) and in *E. glacialis* the form of the alveolus diameter is rounded almost over all their height (Fig. *B*). Epidermal septa delimiting the alveoli on a tangential section on their long sides (in parallel with the long side of alveolar oval) and which are called longitudinal septa, in Mystacoceti (Fig. *A*8) penetrate deeper into the subpapillary layer of the dermis than transversal septa (Fig. *A*9). But in most Odontoceti their height is nearly equal (Figs. *B*8, *B*9). In the epidermis of Odontoceti projections from epidermal septa (Fig. *B*15) grow into the tissue of dermal papillæ (Fig. *B*7). In *D. delphis* and *L. obliquidens* projections are absent, but there exist separate projecting epidermal cells. In Balænopteridæ the epidermal septa have no projections except small ones found in *B. acutorostrata*. In *E. glacialis* projections reach a considerable size. The functional significance of those projections consists in increasing the strength of cohesion between epidermis and dermis and in enlarging the area of the producing surface of epidermis, namely, of its basal layer.

In more cold-loving Balænopteridæ the subpapillary layer of dermis is nearly absent, being replaced by a subcutaneous fat tissue (Fig. *A*3). Fat cells are located immediately under the epidermis, creating thereby a reliable thermoisolation. In most Odontoceti the subpapillary layer of dermis is well marked (Fig. *B*10), a non-adipose layer being usually present therein (Fig. *B*11). A more active mode of life in predatory whales seems to make necessary the presence of a skin layer exposed to a considerable mechanical loading, when pursuing the prey. *D. delphis* and *L. obliquidens*, due to their comparatively small body-size, require a better thermo-isolation than large whales. Therefore, their subcutaneous fat tissue is relatively more developed than in other Odontoceti. *E. glacialis* has a well-developed subpapillary dermal layer with a non-adipose part in it.

The correlation between the dermis and the subcutaneous fat tissue must change according to the seasons of the year, and it is quite possible that in the dermis of all Odontoceti large accumulations of fat cells are formed in winter. We had no samples of the skin from whales obtained late in autumn or in winter, so our description refers to the summer period only.

Characteristic feeding habits of Balænopteridæ are connected with a considerable mobility of their skin, particularly on the ventral side of the body. Owing to this characteristic the net of elastic fibres is strongly developed in their skin. Besides, there are numerous specific bundles of elastic fibres (Fig. *A*17) scattered in the subcutaneous fat tissue. Moreover, in their subcutaneous muscles bundles of striated muscle fibres (Fig. *A*18) are surrounded by numerous elastic fibres (Fig. *A*19) running along them. In Odontoceti the net of elastic fibres in the skin is much less developed, bundles of the same fibres are absent in their subcutaneous fat tissue and there are no elastic fibres in subcutaneous muscles (Fig. *B*18). Active *D. delphis* and *L. obliquidens* possess dense nets of elastic fibres, and at the limit with their subcutaneous muscles plexuses of elastic fibres or even small bundles are to be found. In *E. glacialis* there is an ordinary net of elastic fibres, bundles of the latter being few in the subcutaneous fat tissue.

In subcutaneous muscles of the above-named whale elastic fibres are rare.

In Mystacoceti the skin of the snout has separate profusely innervated sensory hairs. In most Odontoceti there are no hairs in the post-embryonic period, but they are present in embryos. *Monodon* and *Delphinapterus* have no hairs at all even in the embryonic condition.

Besides all the characters indicated above the skin of Balænopteridæ is distinguished by the presence of longitudinal folds on the greater part of the body's ventral side, with the thickest epidermis, its corneous layer, the subpapillary dermal layer and the highest dermal papillæ at the tops of the folds. A gradual decrease in the size of those elements occurs towards the bottom of furrows between the folds.

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[1] Fjelstrup, A., *Zoologischer Anzeiger* (1888). Japha, A., *ibid.*, **29**, No. 14 (1906); *Zoologische Jahrbucher*, **24** (1907), *ibid.* (1910). Kukenthal, V., "Vergleichend-Anatomische und Entwickelungsgeschichtliche Untersuchungen an Waltieren" (Jena, 1889). Sokolov, W., *Bull. Soc. Naturalistes de Moscou*, Section Biologique, **60**, No. 6 (1955); Proc. Fifteenth International Congress of Zoology, London, 277 (1959). Weber, M., "Studien uber Saugetiere. Ein Beitrag zur Frage nach dem Ursprung der Cetaceen" (Jena, 1886).

AN ANTIBODY INHIBITOR

By Dr. JACK G. MAKARI

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IN our outlook on immunology in relation to disease we have specifically concentrated on antigens and antibodies, and we have attempted to explain all humoral host responses in these terms. Clearly there are areas which are not explained by this limited concept. What happens to the host before an antibody is produced ? Does the body lack an early mechanism to tie up an invader before the antibody is formed ? What does the body do in order to rehabilitate its reactive globulins or to stop the production of antibodies when the antigenic stimulus disappears ? Furthermore, we have mounting evidence to show that auto-antibodies directed against the host's own constituents are being formed in even so-called normal humans. How is this auto-antibody handled ? How is it tied up so that it cannot reach its target ? Furthermore, how do we explain psychosomatic influences, hormonal factors, and the impact of stress on the organism in terms of immunology ?

All these factors require that we expand our concept of host defences to include a triad of antigen, antibody, and inhibitor as follows :

$$\begin{tikzcd}
\text{Antigen} \arrow[rr, shift left] \arrow[dr, Rightarrow] & & \text{Antibody} \arrow[ll, shift left] \\
& \text{Inhibitor} \arrow[ur, Rightarrow] &
\end{tikzcd}$$

I have for many years been interested in this mechanism. I have described an inhibitor earlier[1], studied some of its behaviour *in vitro* as demonstrated by its capacity to inhibit flocculation of normal globulins in the presence of cephalin cholesterol emulsion, and investigated its behaviour by means of serial cephalin flocculation curves[2].

Previously I found that immunization resulted in an increase in this inhibiting factor prior to and independent of the specific antibody response. Drugs (such as antimalarials), alcohol, emotional excitement, stress and even physiological mechanisms, such as food and physical exercise[3], all influenced this response. Furthermore, in the study of host-parasite interaction, as occurs in malarial infection, it was apparent that this inhibitor was a resistance factor of vital significance to the host[2].

In this present study evidence is presented to show the dual function of this inhibitor. For this same inhibitor, which can inhibit the flocculation of globulins in the presence of cephalin–cholesterol–ether emulsions, is, in fact, an antibody inhibitor, capable of preventing the fixation of antibodies to uterine segments in the Schultz–Dale test.

In the preliminary experiment shown in Table 1, the source of antibodies for this *in vitro* fixation to uterine segments was the serum from patient *P.*, a case of active melanoma tumour with metastasis, and from patient *B.*, a case of leukoplakia without any evidence of tumour. The two uterine horns (obtainable from a healthy, unsensitized guinea pig) were then removed. Each of these horns was then cut in half into two segments. The resulting four segments were then ready for coating. Two of these uterine horns were immersed in 0·5 ml. of serum diluted with 0·5 ml. of Tyrode solution ; the other two segments were immersed in 0·5 ml. of serum diluted with 0·5 ml. of inhibiting factor. To each of these solutions (to be used for coating) 1·5 ml. Tyrode solution was added. Contact of the serum–Tyrode mixture with uterine segments was done at 0–5° C. (that is, surrounded by crushed ice) for 1 hr. with air bubbled through the mixture throughout the period of contact.

The coated uterine segments were then used in a Schultz–Dale bath[4-6]. The antigens used to release

Table 1. INFLUENCE OF NORMAL INHIBITOR ON FIXATION TO UTERINE SEGMENTS (AS SHOWN BY THE SCHULTZ–DALE TEST) OF ANTIBODIES FROM SERA OF TWO PATIENTS ; ONE WITH TUMOUR AND THE OTHER IN THE PRE-TUMOUR STAGE

Contraction index$_{1,000}$ values are given below

Releasing antigen		Source of serum antibody Pt. *B.*—Leukoplakia				Source of serum antibody Pt. *P.*—Melanoma			
Type	Amount in ml.	Tyrode		Inhibitor		Tyrode		Inhibitor	
		Horn I	Horn II	Horn III	Horn IV	Horn I	Horn II	Horn III	Horn IV
Normal serum	0·5 ml.	0	0	0	0	0	0	0	0
Normal antigen	0·4 ml.	0	0	0	0	0	0	0	0
Carcinoma antigen (supernatant)	0·4 ml.	1,000	1,000	180	0	1,000	830	1,000	1,000
Carcinoma antigen (supernatant)	0·4 ml.	0	260	0	0	0	0	450	590
Melanoma antigen (supernatant)	0·4 ml.	1,000	1,000	0	0	920	920	1,000	1,000
Melanoma antigen (supernatant)	0·4 ml.	1,000	1,000			0	0	1,000	890

Table 2. Effect of Normal Antibody Inhibitor on Fractions from Pooled Sera containing Antibodies to Tissue Antigens, Amino-acids and Other Key Substances
Contraction index values are multiplied by 1,000

	Antibodies to tissue and serum antigens				Antibodies to amino-acids				Antibodies to other key substances			
	No. of tests	*A* Mean C.I. with Tyrode	*B* Mean C.I. with normal inhibitor	*B/A* Ratio of inhibition	No. of tests	*A* Mean C.I. with Tyrode	*B* Mean C.I. with normal inhibitor	*B/A* Ratio of inhibition	No. of tests	*A* Mean C.I. with Tyrode	*B* Mean C.I. with normal inhibitor	*B/A* Ratio of inhibition
Benign 50 per cent fraction	19	102	93	0·91	7	0	0		17	110	52	0·47
Ca. local 50 per cent fraction	22	188	94	0·50	16	290	96	0·33	51	111	36	0·32
Ca. local extension 50 per cent fraction	27	161	50	0·31								
Ca. Metastatic (+ test) 50 per cent	25	162	98	0·60	6	0	0					
Ca. Metastatic (– test) 50 per cent	29	127	54	0·43								
Ca. Cure 50 per cent	20	97	26	0·27								
Ca. local 33 per cent	11	89	154	1·73								
Ca. local extension 33 per cent	18	119	170	1·43								
Ca. Metastatic (+ test) 33 per cent	19	221	105	0·48								
Ca. Local 66 per cent	11	199	35	0·18	7	0	0		12	45	35	0·78
Ca. Local extension 66 per cent	10	18	92	5·11								
Ca. Metastatic (+ test) 66 per cent	24	210	177	0·84								
Ca. Metastatic (– test) 66 per cent	24	431	301	0·70	11	314	224	0·71	15	124	108	0·87

the reactions were supernatant solutions obtained from 20 per cent suspensions of carcinoma and melanoma tissues in isotonic sodium chloride, after removal of the sediments obtained at 14,000 r.p.m. The Tyrode solution used has half-strength calcium and ten times the normal amount of magnesium (sodium chloride, 9·0 gm.; potassium chloride, 0·20 gm.; calcium chloride, 0·05 gm.; magnesium chloride, 0·10 gm. (of crystalline magnesium chloride use 0·21); glucose, 1·00 gm.; sodium bicarbonate 1·00 gm.; sodium dihydrogen phosphate, 0·05 gm.; demineralized distilled water, 1 l.).

A corresponding normal antigen also used to release the reactions was prepared in the same way from several normal tissues (œsophagus, stomach, colon, lung). These normal tissues, which were obtained from a young victim of a car accident at autopsy, were pooled.

The results of this experiment (shown in Table 1) indicate the presence of two types of antibodies to tumour antigens: (1) those antibodies inhibited by normal inhibitor and present in the serum from the leukoplakia case: (2) those antibodies not inhibited by normal inhibitor found in the serum of the patient with active melanoma.

The finding that specific desensitization is possible to carcinoma antigens indicates the antigen–antibody basis of this reaction.

In the other experiments shown in Table 2 sera from 26 individuals with benign and malignant tumours were pooled according to the nature of the tumour, the state of tumour spread and the result of my tumour skin test[7,8] as follows: (1) Benign tumours—tumour skin test +, mean age 39·8 (4 individuals). (2) Carcinoma—early localized—tumour skin test +, mean age 60·3 (3 individuals). (3) Carcinoma—stage of local extension—tumour skin test +, mean age 47·5 (6 individuals). (4) Carcinoma—stage of metastasis—tumour skin test + mean age 54·2 (5 individuals). (5) Carcinoma—stage of metastasis—tumour skin test –, mean age 63 (4 individuals). (6) Carcinoma—cure (survival beyond 5 years)—tumour skin test –, mean age 69 (4 individuals).

These various pooled sera were then fractionated with ammonium sulphate into the following fractions: 33, 50 and 66 per cent. The precipitate in each case was dissolved in isotonic sodium chloride and brought to the original volume of the serum. Each serum fraction was then dialysed against running tap water, distilled water, sodium chloride 1 per cent, and phosphate buffer (sodium chloride, 6·8 gm.; sodium hydrogen phosphate, 0·81 gm.; potassium dihydrogen phosphate, 1·04 gm.; distilled water, 1,000 ml.: *p*H 6·8). The various fractions were then used for *in vitro* coating of uterine segments in the presence or absence of inhibiting factor from a normal blood donor as in the previous experiment.

The inhibiting factor was isolated by the following procedure: to the supernatant after the extraction of 66 per cent fraction, 2*N* sulphuric acid in 50 per cent ammonium sulphate was added slowly with mechanical stirring until the *p*H became 4·8. The precipitate was separated by centrifugation for 45 min. at 10,000 r.p.m. and discarded. The supernatant was then saturated with crystals of ammonium sulphate with continuous mechanical stirring for 1–2 hr. This was left over-night and then centrifuged for 45 min. at 10,000 r.p.m. The supernatant was discarded. The precipitate was dissolved in isotonic sodium chloride solution enough to bring it back to the original volume of serum. It was then dialysed in the same way as the other blood fractions from the diseased sera.

The coated uterine segments were then used in the Schultz–Dale test. To trigger the anaphylactic reactions, tissue and serum antigens obtained from normal and tumorous subjects were used, as well as various amino-acids and other key substances (such as deoxyribonucleic acid, adenylic acid, thymine, purine, pyrimidine, etc.).

Earlier work[9] has demonstrated the presence of antibodies to amino-acids (*a–a* antibodies) and antibodies to other key substances (*k–s* antibodies) in the sera of patients with cancer. An attempt was made in these experiments to see whether the inhibiting factor is effective in inhibiting *a–a* and *k–s* antibodies as well as antibodies to tissue and serum antigens.

The resulting specific anaphylactic reactions were then compared to the contraction of the same uterine segment when 10γ of histamine were added to the bath. This ratio of the specific contraction to that of histamine, that is, the contraction index, was further multiplied by 1,000 to obtain contraction index 1,000.

A ratio of inhibition was obtained by dividing the mean contraction index in the presence of normal inhibitor by the mean contraction index in the presence of Tyrode solution. The effect of inhibiting factor from a normal subject on the fixation to the uterine segments of antibodies, found in the various fractions from pooled sera of patients with tumours, is shown in Table 2. It is apparent from this Table that most of the antibody globulins studied are amenable to the inhibiting action of inhibiting factor (that is, preventing the antibody fixation to the uterine segment) as evidenced by inhibition ratios less than 1·0. Other antibody globulins, however, are not inhibited by inhibiting factor. Instead, a reversal seems to take place so that these antibodies are more fixed to the uterine segments in the presence of inhibiting factor. Thus all the globulins in the 50 per cent fractions could be inhibited as indicated by inhibition ratios less than 1·0 (the lowest value being that of a carcinoma cure pool).

Of the antibody globulins precipitated at 33 per cent only one out of the three fractions tested are inhibited by inhibiting factor (namely, that of carcinoma metastatic, tumour skin test +). The other two 33 per cent fractions from carcinoma local and carcinoma local extension show a reversal of inhibition in the presence of inhibiting factor indicating an abnormality of these latter two antibodies.

Of the antibody globulins precipitated at 66 per cent three out of the four showed inhibition by inhibiting factor (namely, those from fractions studied from carcinoma—local, carcinoma—metastatic tumour skin test +, carcinoma—metastatic tumour skin test −). The fourth serum fraction, representing antibody globulins precipitated at 66 per cent and obtained from the carcinoma—local extension pool, showed a marked reversal of inhibition to inhibiting factor (inhibition ratio being 5·11).

All the *a–a* and *k–s* antibodies studied seem to be amenable to inhibition by inhibiting factor. Again, those associated with the 50 per cent fraction seem to be more amenable to inhibition by inhibiting factor than those associated with the 66 per cent fraction.

These results demonstrate the presence of an antibody inhibitor in normal serum capable of inhibiting the fixation of antibody globulins to smooth muscle in the Schultz–Dale test. This same inhibitor is also capable of inhibiting the flocculation of normal globulins in the presence of cephalin–cholesterol–ether emulsions.

In some stages of cancer, inhibiting factor is capable of inhibiting antibodies associated with certain serum fractions, while in other stages, inhibiting factor is incapable of inhibiting antibodies associated with certain serum fractions. In these latter cases there seems to be a reversal of function so that even a normal inhibiting factor will result in increased, rather than decreased, fixation of antibodies to smooth muscle.

Other than this immunological defect in antibodies, found in certain serum fractions in cancer, there seems to be a defect in the inhibiting factor itself found in cancer sera. Evidence for this finding will be presented elsewhere.

I believe that this new concept of antibody inhibitor will prove to be not only of academic interest but also of assistance in allergic diseases, auto-immune disease and in the study of the ageing process and cancer.

[1] Makari, J. G., *Nature*, 160, 201 (1947).
[2] Makari, J. G., *J. Trop. Med. and Hyg.*, 49, 70 (1946).
[3] Makari, J. G. (unpublished results).
[4] Makari, J. G., *Brit. Med. J.*, 2, 1291 (1955).
[5] Makari, J. G., *Brit. Med. J.*, 2, 358 (1958).
[6] Makari, J. G., *J. Amer. Geriatrics Soc.*, 7, 611 (1959).
[7] Makari, J. G., *Amer. J. Pub. Health*, 49, 687 (1959).
[8] Makari, J. G., *J. Amer., Geriatrics Soc.*, 8, 4, 16 (1960).
[9] Makari, J. G., *J. Geront.*, 14, 494 (1959).

A BIVALVE GASTROPOD

By Dr. L. R. COX, O.B.E., F.R.S., and Dr. W. J. REES
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THE recent discovery of a remarkable mollusc, described as *Tamanovalva limax* gen. et sp. nov. by S. Kawaguti and K. Baba[1], calls for special notice, as this organism, while undoubtedly a gastropod, possesses a bivalve shell ; it is therefore of considerable interest to malacologists who are concerned with the possible origins of bivalve molluscs.

This unique type of gastropod, of which 200 or more specimens have been found, occurs in the sublittoral zone at Bisan Seto on the Inland Sea of Japan. The largest shells are 7 mm. long. The main features of the animal are :

(1) A slug-shaped head–foot mass which can be protruded from the shell and extends well to the front of it when the animal is crawling ; it can be completely withdrawn between the two valves, which close as in a lamellibranch. The foot is narrow and has a flat sole which can be folded longitudinally along the median line.

(2) Two stout rhinophores and two small lobiform oral tentacles on the well-developed head, and eyes which lie close together on a small median projection some way from its extremity.

(3) A radula of the sacoglossan type, in which only one tooth is in operation at a time, when the animal is feeding on its host plant (the alga *Caulerpa*).

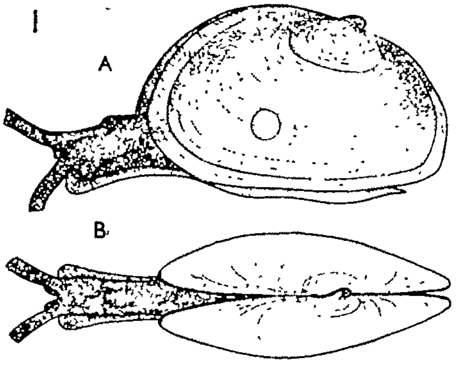
(4) A male genital aperture with penis, located far forward on the right side of the body, just behind the head.

(5) A bilobed mantle open along the ventral margins.

(6) A single gill, formed of a series of thin lamellæ as in the sacoglossan genera *Oxynoe* and *Lobiger*, and situated under the right mantle lobe.

(7) The anus opening into the mantle cavity well to the rear.

These structures and others that need not be mentioned are indicated in Figs. 1 and 2. There are no



Figs. 1*a*, *b*. *Tamanovalva limax* *a*, lateral view from left; *b*, dorsal view: note the rhinophores, eyes and protoconch (× *c*. 6·6) (After Kawaguti and Baba)

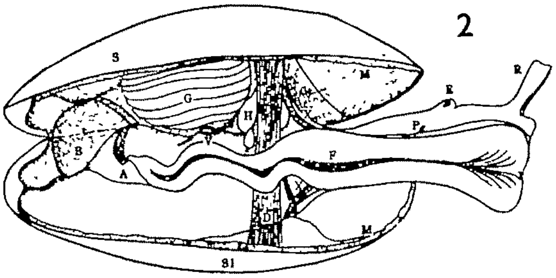


Fig. 2. *Tamanovalva limax*. ventral view (× 8). *A*, anus; *B*, hypobranchial gland; *D*, adductor muscle; *E*, eye; *F*, foot; *G*, gill; *H*, heart; *M*, mantle edge; *P*, penis pore; *R*, rhinophore, *S*, right valve of shell; *Sl*, left valve of shell, *V*, vaginal pore; *X*, head retractor muscle. (After Kawaguti and Baba)

siphons and no parapodia. The nervous system is not described.

The most remarkable feature is the shell, consisting of two valves which are equal in size and shape except for a small helicoid spiral structure at the apex of the left valve. There is a single well-developed adductor muscle which connects the two valves of the shell slightly in front of the middle of its length. The valves are joined along the hinge-line by an 'invisible' (? transparent) ligament; but there are no hinge-teeth.

Eggs are laid in sausage-shaped masses about 10 mm. long. The developing embryo passes through trochophore and veliger stages, and when the veliger hatches out it has a bilobed velum, a helicoid shell and an operculum. It is the larval shell that is preserved, as a protoconch, at th eapex of the left valve of the adult shell; it appears to be sinistrally coiled although the animal is dextrally organized (a feature of the typical opisthobranch protoconch)

The authors conclude that *Tamanovalva* is an opisthobranch gastropod belonging to the order Sacoglossa, as shown particularly by its slug-like body with two stout rhinophores and the nature of its radula. In view of its remarkable bivalve shell they have placed it in a new suborder Tamanovalvacea.

That this animal is a gastropod is indisputable, and its reference to the order Sacoglossa seems well justified. The opisthobranchs are now considered to be the most advanced sub-class of the Gastropoda, and many have contrived to counteract the disadvantages of torsion and asymmetry possessed by their prosobranch ancestors by the acquisition of secondary symmetry, of which *Tamanovalva* surely provides a further and most striking instance. It is to be interpreted as an evolutionary experiment in a sub-class not represented among the earlier known gastropods and in an order of which no fossil representatives have yet been recognized. The mere fact, however, that it demonstrates the possibility of a bivalve and almost symmetrical shell becoming developed in a normally univalve group lends support to the theory that the Bivalvia (Lamellibranchia) were derived originally from primitive univalve molluscs. Whereas, however, it has been supposed that this event involved the bending and hinging of a symmetrical, cap-like shell along its median line and the fusion of pallial muscles to form the two adductor muscles[2], the origin of the second valve and of the adductor muscle of *Tamanovalva* cannot be satisfactorily discussed until the post-embryonic ontogeny of this form has been investigated. It would appear that the protoconch-bearing left valve represents the original univalve shell and that the second valve is an extension which presumably arises by bilobation of the mantle at a relatively early stage in ontogeny (probably at the time of metamorphosis) and becomes hinged. Being quite independent of the foot, it clearly cannot arise from the operculum, which presumably, as in most other opisthobranchs, atrophies at an early stage in development.

Kawaguti and Baba refer briefly to *Edentellina* Gatliff and Gabriel[3], a supposed lamellibranch which, like their new genus, is characterized by the presence of a small coiled larval shell at the tip of one valve; but they consider it to be quite distinct. We are by no means convinced about this. The type-species of *Edentellina*, *E. typica*, was first discovered along the coast of Victoria and has since been found off South Australia and Western Australia. A second species, *E. corallensis*, dredged off Queensland within the Great Barrier Reef, has been described by Hedley[4], who at the same time has also re-described the type species. It seems significant that, according to that author, *E. typica* "lives in shallow water a little below the level of low tide" and that he had "commented on the absence from *Edentellina* of characteristic pelecypod features and suggested that possibly it might be the internal shell of a tectibranch" (tectibranchs are opisthobranch gastropods).

Hedley's figures of *E. typica*, which are more satisfactory than the rather poor photographs that illustrated the original description of the species, represent a small shell closely resembling *Tamanovalva* in shape. Although identified as a right valve from its outline (on the assumption that it belonged to a lamellibranch), the valve bearing the coiled larval shell at its tip is very similar to the corresponding valve (known to be the left one) of the Japanese form. In the original description of *E. typica* the shell was described as being without hinge-teeth, but afterwards, when better specimens were obtained, a small tooth-like structure was detected in both valves. Similar but possibly more obscure structures may have been overlooked in the Japanese species. The position of the adductor scar or scars has not been observed in *Edentellina*, while Hedley's statement that the soft parts of the animal are unknown appears still to hold good. It is, therefore, now all the more desirable that a special search for living specimens should be made in Australian waters, to ascertain whether or not this is a gastropod genus synonymous with *Tamanovalva*.

There is a further point, of special interest to palæontologists. As pointed out in Hedley's paper, a rare species found fossil in the Eocene of the Paris Basin appears to be a representative of *Edentellina*. It was known to Deshayes, who had assigned the

MS. name *Ludovicia squamula* gen. et sp. nov. to it, but was first described by Cossmann in 1888[5], by which time the name *Ludovicia* had already been published in connexion with an entirely distinct organism. The Paris Basin fossil is a small, thin-shelled bivalve of the same general trapeziform shape as *Edenttellina* and *Tamanovalva*, and young specimens have, at the tip of the left valve, a small spiral larval shell which is later lost. Although he included it in the lamellibranch family Galeommidae, Cossmann remarked, "j'aurais pu croire qu'il s'agissait d'un gastropode". If it is accepted as an *Edenttellina*, and if the latter proves to be identical with the newly described Japanese genus, we shall know for the first time that the opisthobranch order Sacoglossa existed so long ago as the Eocene.

The authors' full report on the remarkable aberrant form described in their paper will be eagerly awaited.

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[1] *Biol. J. Okayama Univ.*, **5**, 177 (1959).

[2] Yonge, C. M., *Trans. Roy. Soc. Edin.*, **62**, 444 (1953).

[3] *Proc. Roy. Soc. Victoria*, N.S. **24**, 190, pl. 46, Figs. 5, 6 (1911).

[4] *Proc. Malac. Soc. Lond.*, **14**, 76 (1920).

[5] *Ann. Soc. Roy. Malac. Belg.*, **22**, 45, pl. 2, Figs. 21, 22.

THE SECONDARY PAIRING OF BIVALENTS WITH GENETICALLY SIMILAR CHROMOSOMES

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IN some allopolyploid plants, at first metaphase of meiosis, the bivalents are not randomly distributed but occur together in pairs or groups. This secondary pairing has been assumed to take place between bivalents composed of genetically similar pairs of chromosomes[1,2], but this assumption has apparently never been tested. Material suitable for such a test can, however, now be produced in the allohexaploid common wheat (*Triticum aestivum* $2n = 6x = 42$), in which secondary pairing may be occasionally observed.

The material used in the present investigation was developed from three parental lines in the variety Chinese Spring, generously supplied by Dr. E. R. Sears. These had chromosomes III, XII and XVI, in turn, represented by a pair of telocentric chromosomes for one arm, the other arm being completely deficient. Chromosomes III, XII and XVI are homœologous[3], that is, genetically equivalent chromosomes, belonging respectively to the *B*, *A* and *D* genomes derived from the original diploid ancestors of wheat[4].

The three lines were crossed together in pairs to produce *F*1's with one normal and one telocentric chromosome for each of two pairs. Thus, in the cross between the line with chromosome III telocentric and the line with chromosome XII telocentric, the hybrid had one telocentric member of the III and of the XII pair, and one normal member of each pair.

Meiotic pairing between the normal and telocentric chromosomes led to the formation of heteromorphic, rod-shaped bivalents, since chiasma-formation could take place only in one arm. The heteromorphics were readily recognizable in meiotic preparations, so that the relative positions of the two bivalents at first metaphase could be recorded.

The preparations analysed were Feulgen-stained squashes of pollen mother cells from anthers fixed acetic alcohol. Only cells having strictly linear arrangements of chromosomes were scored (Fig. 1), and the scoring recorded the number of bivalents which intervened between the two recognizably heteromorphic pairs. This method is artificial in that a structure which was originally two-dimensional, the metaphase plate, is represented as one-dimensional. However, it seems unlikely that squashing resulted in systematic distortions of the relative spacings of bivalents on the original plates. Moreover it should be indicated that the score of the number of bivalents intervening between the heteromorphic pairs is merely a measure of the degree of separation, and does not represent the number of bivalents by which they were separated on the original plate. With these reservations, however, the technique of recording seems satisfactory.

The data derived from scores of the three types of *F*1's with two heteromorphic bivalents are recorded in Table 1. There are no significant differences between the three distributions (III/XVI – III/XII, $\chi^2_{(14)} = 24{\cdot}56$, $P = 0{\cdot}1$–$0{\cdot}5$; III/XVI – XII/XVI, $\chi^2_{(15)} = 19{\cdot}03$, $P = 0{\cdot}3$–$0{\cdot}2$; and XII/XVI – III/XII, $\chi^2_{(14)} = 18{\cdot}86$, $P = 0{\cdot}2$–$0{\cdot}1$), so the data have been summed. The summed distribution falls off steeply from a clear maximum of cells with the heteromorphic bivalents adjacent, to minimum frequencies with the heteromorphics at opposite ends on the plate. The observed distribution has been tested against that expected from the random alignment of bivalents. This may be calculated from the generalized formula $\frac{2(n-1-r)}{n(n-1)}$, where n is the total number of bivalents and r is the number of intervening bivalents, for which I am indebted to Dr. W. F. Bodmer. The random distribution is a straight line which in the present instance runs from a frequency of 0·0952

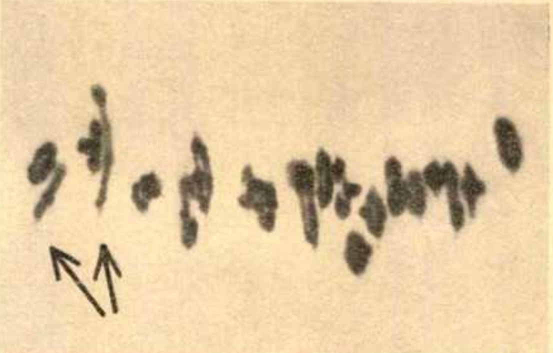

Fig. 1. First metaphase of meiosis in a plant of *T. aestivum* with one telocentric member of both the chromosome III and XVI bivalents. There is one intervening bivalent between the heteromorphic bivalents III and XVI, which are arrowed. (× 1,250)

Table 1. NUMBER OF BIVALENTS SEPARATING HOMŒOLOGOUS PAIRS IN FIRST METAPHASE CELLS

No. intervening bivalents	Cells per intervening class					χ^2
	III+ XII marked	III+ XVI marked	XII+ XVI marked	Total	Random expectation	
0	21	40	23	84	39·22	62·55
1	9	21	15	45	37·29	1·59
2	8	15	12	35	25·30	1·12
3	9	10	8	27	33·33	1·20
4	6	10	6	22	31·39	2·81
5	6	11	8	25	29·42	0·66
6	6	9	6	21	27·48	1·52
7	6	8	5	19	25·50	1·66
8	9	3	5	18	23·57	1·32
9	9	11	5	25	21·59	0·54
10	5	4	1	10	19·61	4·71
11	6	4	5	15	17·67	0·40
12	4	3	4	11	15·69	1·40
13	5	3	3	11	13·72	0·54
14	3	6	1	10	11·78	0·27
15	4	4	3	11	9·81	0·14
16	3	3	6	12	7·87	2·17
17	1	2	2	5	5·89	
18	3	4	1	8 } 19*	3·96 } 11·83*	4·35
19	0	3	3	6	1·98	
Total	123	174	122	412	412·00	88·95

$\Sigma\chi^2_{(17)} = 88·95$; $P = < 0·001$

* Classes summed to increase the value of the expected.

with no interveners to 0·0048 with 19 interveners (Fig. 2).

There is a marked and highly significant deviation of the observed distribution from random ($\chi^2_{(17)} = 88·95$, $P = < 0·001$), and the largest single deviation is in the class with no intervening bivalent. Indeed, when the ratio of observations of the combined classes 1 to 19 : 0 is tested against random, the deviation is highly significant ($\chi^2_{(1)} = 56·51$, $P = <0·001$). Thus the major disturbance of the random distribution results from the heteromorphic bivalents occurring together too frequently.

If this represents secondary pairing, in the present experimental situation the apparent attraction between the marked homœologues may well have been affected by the deficiency of an arm from the telocentric member of each pair. If genetically corresponding arms were missing from the marked pairs, the amount of structurally similar material would be reduced, depending on the relative lengths of the arms of each chromosome, by about a quarter. Similarly, if the arms missing in each pair were not genetically equivalent, there would be only half the normal amount of structurally similar material. Consequently, if pairing depends on the degree of structural similarity, results based on pairs with telocentric members may under-estimate normal attraction.

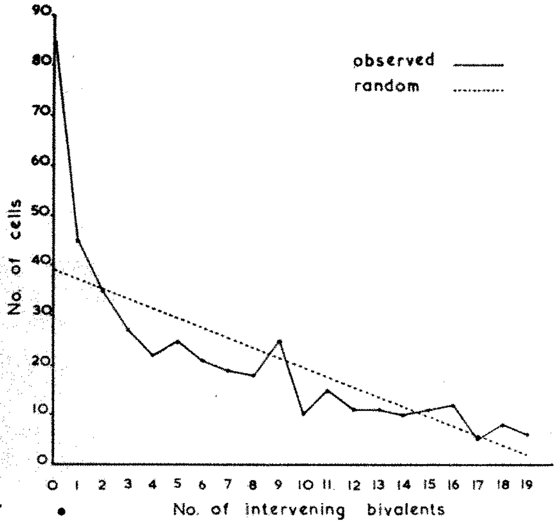

Fig. 2. Observed and random distributions of cells with the heteromorphic homœologous bivalents separated by 0–19 unmarked bivalents

Also the degree of secondary pairing might have been diminished because the marked bivalents were always rods, and rod bivalents probably move less freely than rings during the metaphase congression. Moreover, wheat is a hexaploid in which secondary pairing is ideally possible between all three bivalents involving chromosomes III, XII and XVI. However, in the material so far available, only two of these pairs have been simultaneously recognizable ; consequently the influence of the unmarked pair could not be determined. The excess of the class with one bivalent intervening may, however, be due to the separation of the marked pairs by their unmarked homœologue.

The excess of the combined classes with 17, 18 and 19 interveners may reflect a slightly non-random component in the arrangement of rod bivalents. Possibly they were more often at the periphery of the original plate, or their positions may have influenced the orientation of the plate on squashing. However, the majority of adjacent heteromorphic bivalents were in the interior rather than at the ends of the plates, so that this factor has probably not greatly influenced the recorded frequency of adjacent pairs. In subsequent work with marked non-homœologous as well as homœologous pairs, and in material with all three homœologues marked, this problem may be clarified.

Nevertheless, despite the intrusion of other disturbance, the considerable excess of immediately adjacent homœologous pairs seems to be reasonable evidence that secondary pairing brings genetically similar bivalents together, as had been supposed. If this is so, homœologous pairs must be attracted together following the wide dispersal of bivalents throughout the nucleus during diakinesis. However, the chances of secondary association very likely depend upon the relative proximities of homœologues during diakinesis. In wheat there are probably zygotene attractions between all six homœologous chromosomes but, in the presence of chromosome V, there is complete synapsis only between homologues[5]. If as a consequence of zygotene attractions homœologous bivalents are neighbours in early prophase, they are not likely to be so widely separated at diakinesis as non-neighbours, so that secondary pairing at metaphase will readily follow.

Evidence of secondary pairing between bivalents III, XII and XVI is a useful supplement to the demonstration of similarity in their genetic activity[3]. It is a further indication of the evolutionary proximity of these three chromosomes, and indeed of the genomes of which they are part. The extent of genetical and structural triplication of chromosomal material of wheat is thus re-emphasized in a way which re-asserts the significance of the genetic regulation causing wheat to form only bivalents, and no multivalents, at meiosis.

[1] Darlington, C. D., *J. Genet.*, **19**, 215 (1928).
[2] Lawrence, W. J. C., *Cytologia*, **2**, 352 (1931).
[3] Sears, E. R., *Res. Bull. Mo. Agric. Exp. Sta.*, 572 (1954).
[4] Okamoto, M., *Wheat Inf. Service*, **3**, 6 (1957).
[5] Riley, R., and Chapman, V., *Nature*, **182**, 713 (1958).

LETTERS TO THE EDITORS

PHYSICS

Surface-controlled Bulk Conductivity in Organic Crystals

In an earlier paper[1] experiments were described which demonstrated hole injection from unexcited and optically excited electrode layers into an adjacent anthracene crystal, resulting in a large increase in the current passing through the bulk of the crystal. In the present communication, the interaction between an optically excited anthracene crystal and its adjacent electrode is reported.

The experimental techniques used have been described in detail elsewhere[2,3]. The general procedure was to use an electrolyte as the electrodes for thin (10μ) single crystals of anthracene. The incident light passed through the electrolyte before striking the crystal face. The current through the crystal was measured with $1M$ sodium iodide and $1M$ sodium chloride solutions in contact with the illuminated side of the crystal, using 3650 Å. light which was transmitted by the electrolyte but completely absorbed by the anthracene.

Table 1. Photoconductivity in Anthracene using Sodium Chloride and Sodium Iodide Electrolyte Electrodes

Applied voltage	(1) Side *A* : NaI *B* : NaI Dark current	(2) *A* : NaI *B* : NaCl Dark current		(3) *A* : NaI *B* : NaI Photocurrent		(4) *A* : NaI *B* : NaCl Photocurrent		(5) *B* : NaI *A* : NaI Photocurrent		(6) *B* : NaCl *A* : NaI Photocurrent	
	$i+ = i-$*	$i+$	$i-$	$i+$	$i-$	$i+$	$i-$	$i+$	$i-$	$i+$	$i-$
(Volts)	× 10^{12} amp.	× 10^{12} amp.		× 10^{9}† amp.		× 10^{9}† amp.		× 10^{9}† amp.		× 10^{9}† amp.	
2				10	0·33	6·5	2·1	4·1	0·40	13·6	0·25
4	0·3	0·2		21	0·40	14·5	3·5	11·5	0·52	41·6	0·40
8	0·5	0·5		29	0·46	22·5	4·3	17·5	0·63	148	0·60
15	0·9	1·1		36	0·52	29	4·5	21·5	0·72	240	0·80
30	1·7	2·5	3·5	40	0·58	33	4·7	24·0	0·81	312	1·05
50	3·0	4·4	6·0	42	0·62	35	5·0	25·2	0·87	328	1·15

Area of crystal, 0·1 cm.^{2}. Intensity of 3650 Å. light, 320μ watt/cm.^{2}. All solutions are 1 M in concentration.
* $i+$ and $i-$ refer to currents measured when the polarity of the side illuminated by light was positive and negative respectively.
† Note change in scale.
The side illuminated by the light is listed at the top of each column in the table.

The experimental results are given in Table 1. The most interesting result was that the magnitude of $i+$ when the illuminated side of the anthracene was in contact with sodium chloride solution (column 6) was 13 times that of $i+$ when this side was in contact with sodium iodide solution (column 5). The current $i-$ did not change much.

It was also noted that the ratio $i+/i-$ in the case shown in column 6 was 285 as compared with a ratio of 7 in the case shown in column 4. The large value of $i-$ in column 4 was felt to be due in most part to an effect of fluorescent light on the anthracene–sodium chloride electrode face not directly illuminated. Scattering effects were ruled out by shielding the unilluminated face. Using identical sodium iodide electrodes, the ratio $i+/i-$ at 50 V. was 68 (column 3). The latter experiment was repeated with no change other than shifting the illumination from one face of the crystal to the opposite face. The ratio $i+/i-$ was 29 (column 5), indicating some crystal asymmetry. The change from sodium chloride to sodium iodide in the electrolyte at the unilluminated side decreases the $i-$ current, however, by a factor of 8, whereas the $i+$ current is almost unchanged (compare columns 3 and 4).

This dependence of $i-$ on the nature of the electrolyte in contact with the crystal face indicates that at least part of the current measured with the illuminated side negative is due to an interaction at the unilluminated electrode which produces positive holes at that face. The total $i-$ current would, therefore, be made up of the flow of these holes from the face not directly illuminated as well as electrons from the illuminated face. On this basis, the flow of electrons in $i-$ in the case shown in column 6 must be less than 0·3 per cent of $i+$.

In all cases, the photocurrents were almost saturated at 50 V. or lower. The dark current did not exhibit saturation.

The quantum efficiency for the photocurrent when the illuminated face of the crystal was in contact with sodium chloride solution was one carrier liberated into the anthracene crystal for thirty quanta falling on the face of the anthracene. In the case where the illuminated face was in contact with sodium iodide solution, the quantum efficiency was one carrier liberated into the anthracene for 390 quanta falling on the face of the anthracene.

In general, these results may be understood by assuming that the observed photocurrents are a result of the creation of an exciton by the light absorbed in the anthracene and its subsequent diffusion to the adjacent electrode, where it undergoes a dissociation into an electron and a positive hole. The latter then moves through the anthracene under the action of the electric field. Any event which will quench the exciton or inhibit its subsequent dissociation will diminish the photocurrent. Apparently such quenching occurs at the electrode by the I^- and its quenching action is greater than that of the Cl^-.

A more complete discussion of these results will be given shortly.

The work of Mr. Peter Gordon, who made most of these measurements, is greatly appreciated.

Grateful acknowledgment is made of the assistance of the Office of Naval Research (*NONR* 285(41)) in this work and of the Air Force Cambridge Research Center, Contract *AF*19(604)–3888.

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[1] Kallmann, H., and Pope, M., *J. Chem. Phys.*, **32**, 300 (1960).
[2] Kallmann, H., and Pope, M., *Rev. Sci. Instr.*, **29**, 993 (1958).
[3] Kallmann, H., and Pope, M., *Rev. Sci. Instr.*. **30**, 44 (1956).

Emission Spectroscopy with Infra-Red Double-Beam Spectrophotometers

SOME time ago, K. A. Fischer and G. Brandes[1] demonstrated that it is possible to obtain useful emission spectroscopic data with an infra-red double-beam spectrophotometer, if the sample placed in the beam path of the spectrophotometer is warmed up with a closed sample beam shutter. Instead of the quotient of the radiation intensity passed by the object and of the unattenuated radiation intensity, the instrument now scans the quotient of the inherent radiation intensity of the object and of the reference beam intensity.

The sensitivity of this method can be increased 100 times, if the intensity of the reference beam is attenuated by means of a Venetian shutter by the same factor. Thus it is possible to record also the relatively weak emission maxima of moderately heated gases.

Fig. 1 shows the emission spectrum of ammonia, heated up to 160° C., between 9 and 12μ, recorded with a ten-fold reference beam attenuation. The thickness of the ammonia sample was 5 cm. The spectrum was recorded with a Model 21 Infra-red Spectrophotometer of Bodenseewerk Perkin-Elmer and Co., GmbH. The attenuation of the reference beam was effected by a reference beam attenuator designed as an accessory to this instrument, the object being warmed up in a heatable gas cell, which likewise constitutes a usual spectrophotometer accessory though serving other purposes otherwise. The spectral dispersion was effected with a sodium chloride prism; the slit-width was controlled with the 'slit programme 800'. This slit programme provides a slit-width of 55μ at a wave-length of 10μ. Because of the low energy striking the thermocouple, the instrument was operated with maximum amplification, maximum time constant and a scanning speed of 5×10^{-4} μ/sec. The spectrum distinctly shows the emission maxima assigned to the absorption bands of the ammonia. Fig. 2 shows the absorption spectrum of the same sample, which was scanned with the same slit programme, however, without reference beam attenuation. It becomes obvious that the frequency conformity of the emission and absorption maxima has been realized quite well. The intensity ratios between the individual maxima are, however, considerably changed.

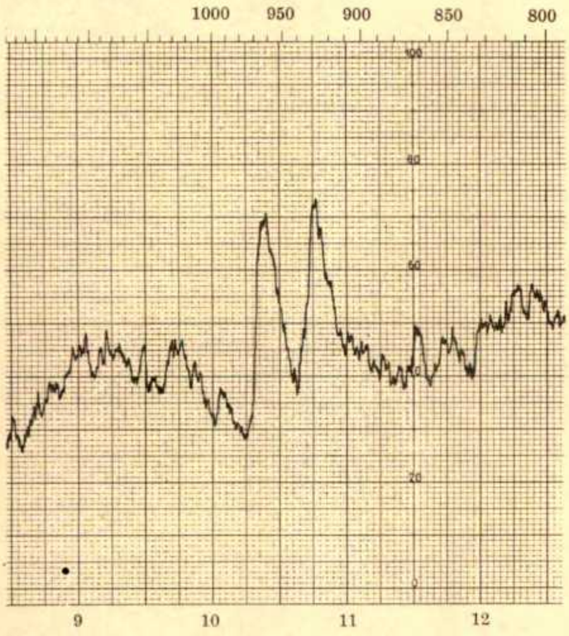

Fig. 1

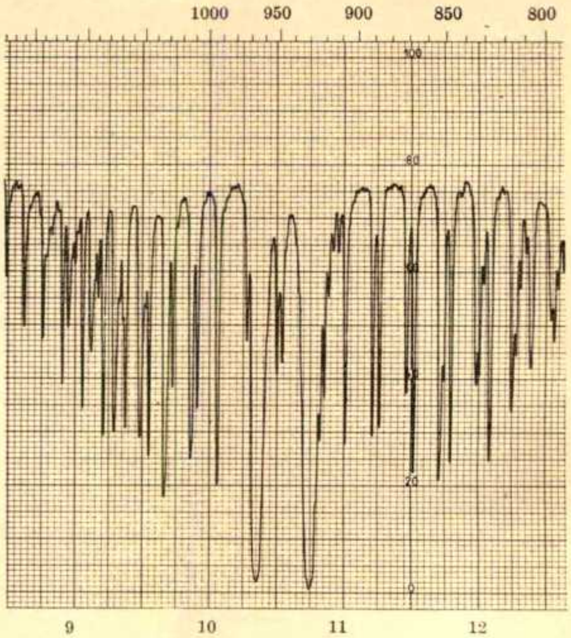

Fig. 2

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[1] Fischer, K. A., and Brandes, G., *Naturwiss.*, **43**, 223 (1956).

Rotational Analysis of the $C^2\Pi - X^2\Sigma^+$ System of CaCl

ALTHOUGH many of the electronic band-systems of the monohalides of Group II*A* are readily observed in emission and absorption, little is known about the properties of these molecules, other than their force constants and the positions of their lower-lying electronic energy states. Only one complete rotational analysis, of the system $A^2\Pi_1 - X^2\Sigma^+$ in BeF has yet been given[1].

In many cases, the rotational structure is, through the near equality of B-values, too close to be resolved, but new photographs on a 6·65-m. grating spectrograph suggest that analyses of some systems will be possible. One such system is $C^2\Pi - X^2\Sigma^+$ in CaCl, which may conveniently be observed in absorption. We have been able to analyse the 0,0 band, and the shorter wave-length sub-bands of the 1,0 and 0,1 bands. The upper state is sufficiently close to Hund's case *a* for the two sub-bands to be treated independently to give effective values of B' and D'. The Λ-type doubling, which is negligibly small in the shorter wave-length sub-bands, but observable in the longer wave-length components, shows that the C state is $^2\Pi_{\text{reg.}}$, presumably $\cdots (w\pi)^4(v\pi)$, $^2\Pi_r$. An earlier, incomplete analysis[2] of this system is incorrect. The constants obtained in the present analysis are summarized in Table 1.

Table 1. SUMMARY OF CONSTANTS FOR THE $C^2\Pi_r - X^2\Sigma^+$ SYSTEM OF $Ca^{35}Cl$

State	T_0	$\Delta G_{0,1}$	B_0	10^4	$10^7 D$	r_e, Å.
$C^2\Pi_{3/2}$	26,557·80	333·86	0·14268	7·47	$1{\cdot}0_0$	$2{\cdot}516_0$
$C^2\Pi_{1/2}$	26,481·82		0·14216		$1{\cdot}0_3$	
$X^2\Sigma^+$	0	367·53	0·15156	$7{\cdot}8_2$	1·0	$2{\cdot}439_0$

It is of some interest to compare the internuclear distances in the ground-states of the monohalides with those found in the dihalides[3]. Figures are given in Table 2. They suggest that the distances in the monohalides are about 3 per cent shorter than in the corresponding dihalide.

Table 2. INTERNUCLEAR DISTANCES (REFS. 1 AND 3)

BeF_2	1·40 ± 0·03 Å.	BeF	1·361 Å.
$BeCl_2$	1·75 ± 0·02	BeCl	(1·7)
MgF_2	1·77 ± 0·02	MgF	$1{\cdot}75_2$
CaF_2	2·10 ± 0·03	CaF	(2·02)
$CaCl_2$	2·51 ± 0·03	CaCl	2·439

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[1] Herzberg, "Molecular Spectra and Molecular Structure", 1, "Spectra of Diatomic Molecules" (Van Nostrand, New York, 1950).

[2] Parker, *Phys. Rev.*, **47**, 349 (1935).

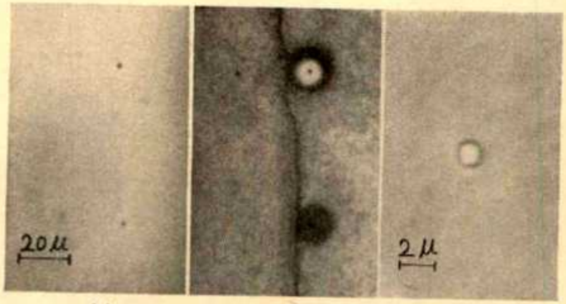
[3] Akishin, P. A., *et al.*, see Bastiansen and Lund, "Ann. Rev. Phys. Chem.", **10**, 43 (1959).

Dark Blue Ring developed by Electron Bombardment in Rock Salt

WE have recently studied the effect of electron bombardment on rock salt crystal obtained from the Halle Company in Germany. It was found that the blue or colloidal coloration was produced on the surface of the specimen bombarded with electrons at high temperature[1]. In the course of the experiments we have also observed that the dark blue rings or circles were scattered on the coloured surface. In this communication we shall be concerned mainly with the behaviour of these rings or circles.

Bombardment was performed as follows. The cleaved specimen was annealed for 60 min. at elevated temperature *in vacuo* and then bombarded with electrons for 15 min. at the same temperature. The bombarding voltage and current density were 18 kV. and 80 μamp./cm.² respectively.

Figs. 1*a* and *b* show the same field of vision of the specimen before and after bombardment, and Fig. 1*c* gives a magnified picture of the small point shown in Fig. 1*a*. These observations show that the rock salt as received contained a large number of small cubic cavities within the crystal (about 10^7/c.c.); and the centre of the dark blue ring corresponds to a cavity just beneath the surface and the centre of the circle to one which is somewhat deeper in the crystal. The edges of this cavity are parallel to the {100} planes of rock salt.



(*a*) (*b*) (*c*)

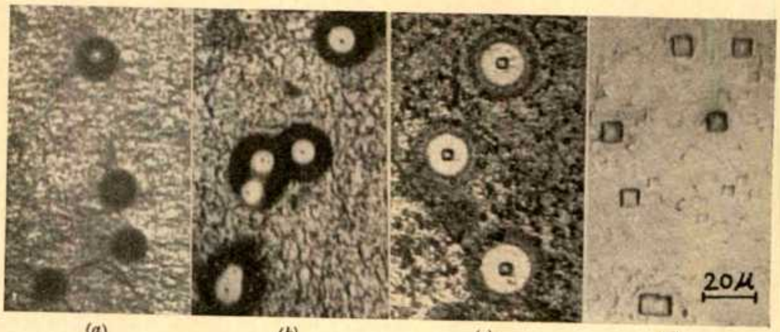
Fig. 1. Dark blue ring and circle developed by electron bombardment. (*a*) Before bombardment; (*b*) after bombardment (*T* = 300° C.); (*c*) magnified photograph of the small point in (*a*)

The changes in the ring pattern are shown in Fig. 2 as a function of temperature during the bombardment. The diameters of both outer and inner circles of the rings increased with the rise of temperature, and at the same time, the dimension of each cubic pit at the centre also increased. At temperatures above 370° C., only large cubic pits were produced.

Further heat treatment of the ring after bombardment led to increase of the inner diameter with the rise of temperature, but the outer ring did not increase, and at about 450° C. it was bleached out entirely.

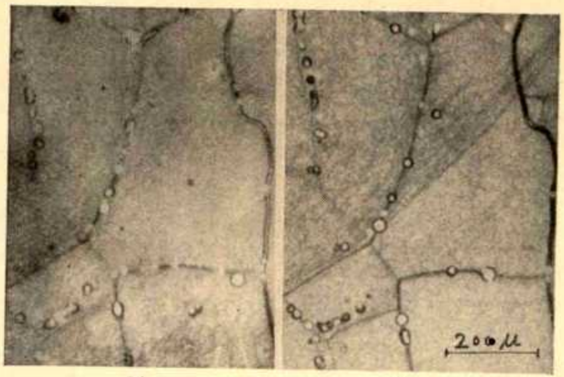
Apart from the dark blue ring or circle, we also observed the development of a coloured contour line as shown in Fig. 1*b*. Figs. 3*a* and *b* show matching surfaces of the cleaved specimen after bombardment; it can be seen that the matching of the contour lines was very good. Hence these contour lines are believed to indicate the sub-grain boundaries.

We are now investigating the mechanism of development of these peculiar types of coloration in



(*a*) (*b*) (*c*) (*d*)

Fig. 2. The change in the ring pattern as a function of temperature: (*a*) 290° C., (*b*) 320° C., (*c*) 360° C., (*d*) 380° C.



(*a*) (*b*)

Fig. 3. Matching surfaces of cleaved specimen after bombardment (*T* = 320° C.); (*b*) was reversed in printing

rock salt. A detailed discussion of the results will be published later.

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[1] Yoshida, S., and Ikeda, T., *J. Phys. Soc. Japan*, **14**, 473 (1959).

CRYSTALLOGRAPHY

Crystal Structure of Mellitic Acid

THE only published crystallographic data for mellitic acid, to the best of our knowledge, are those reported by Deverin[1]. According to his observations mellitic acid is dimorphic: rhombohedral when crystallized from aqueous solution at room temperature, and of an unidentified lower symmetry class when separated from hot aqueous solution.

We obtained large prism-shaped crystals from a hot solution in 50 per cent nitric acid. The crystals were orthorhombic, optically negative, elongated in *b*-axis direction, pseudo-hexagonal, with well-developed forms {100}, {102}, {010}, exhibiting a perfect cleavage parallel to (010). The crystals were not stable for long periods, especially not when irradiated by X-rays, but became opaque and eventually disintegrated in a white crystalline powder. We assumed at first that the loss of nitric acid as solvent of crystallization might be the cause of the disintegration, but the micro-combustion analysis showed that our specimens had been pure mellitic acid. The phenomenon has been, therefore, explained as a transition from the orthorhombic high-temperature modification to another of low temperature, probably identical to the rhombohedral modification, observed by Deverin. It is rather interesting that our orthorhombic crystals, obtained from nitric acid solution, were considerably more stable while in contact with the mother liquor. Therefore, all our X-ray diffraction measurements were carried out with the specimen sealed over a drop of the mother liquor in a Lindeman glass capillary.

The unit cell and space group were determined from the oscillation and Weissenberg diffraction photographs taken with copper $K\alpha$-radiation. The orthorhombic unit cell, which has the dimensions: $a = 16 \cdot 60$ Å.; $b = 8 \cdot 17$ Å.; $c = 19 \cdot 17$ Å., contains eight molecules $C_{12}H_6O_{12}$. The space group is $C^{10}_{2h} - Pccn$. All possible $0kl$, $k0l$ and $hk0$ reflexions were recorded with the help of an integrating Weissenberg goniometer. The relative intensities were determined from the multiple film photographs with the use of a microdensitometer. The ratio of the weakest to the strongest intensity of 1 : 16,000 was obtained.

The Patterson projection on the (010) indicated: (i) that the nolecules of mellitic acid formed layers parallel to the (010), (ii) that in the *b*-axis period two such mutually displaced layers were involved. The overlapping of these layers in the projection gave a vector map with an intricate hexagonal pattern which could not be interpreted satisfactorily. The arrangement of the molecules was then derived from the (100) and (001) Patterson projections. The molecules occupy positions on the diad axes so that the unit cell contains two symmetrically independent groups of four molecules which are defined in the International Tables[2] by the (*c*) and (*d*) sets of co-ordinates, respectively. The ambiguity in the orientation of the molecules toward the diad rotation axis was solved with the help of a trial-and-error procedure in which both the (010) vector map and the $h0l$ intensity data were considered. In this stage of the analysis it became evident that numerous possibilities for locating oxygen atoms would require tedious computational work of successive refinement. Therefore, a new direct method for the phase determination was elaborated by one of us[3] and applied to the sign determination of the structure factors $F(hk0)$ and $F(0kl)$. The electron density maps, computed on the basis of these signs, gave the first co-ordinates of oxygen and carbon atoms, which enabled an efficient refinement of the (010) electron density map, resulting in the complete elucidation of the structure. The final electron density map on the (010) is given in Fig. 1.

All carbon atoms of a molecule lie in a plane which is inclined to the (010) at not more than about two degrees. Each molecule is surrounded by six nearest molecules with the approaches in the direction of their carboxyl groups at the distance of $2 \cdot 62 \pm 0 \cdot 01$ Å. between oxygen atoms. This value for the oxygen separation between two carboxyls is evidence of a strong hydrogen bonding which links molecules together in an endless layer parallel to (010). A preliminary investigation by infra-red spectra confirmed roughly this hydrogen bond-length (Hadži, D., private communication). The next layer is derived in the same way but, in relation to the former, it is displaced along the *c*-axis by the amount of $z_2 - z_1 = 0 \cdot 151$, where z_1 and z_2 are the parameters of the centre of the two symmetrically independent molecules. The whole crystal structure of mellitic acid is thus built up by the alternation of these two layers with the separation of $1/2b$ between them (Fig. 2). Since the diad axis lies in the plane of the benzene ring and passes through two opposite carboxyl carbon atoms, the orientation of six carboxyls of one molecule is defined by four planes differently inclined to the plane of the benzene ring. The most inclined pair of carboxyls approaches the carboxyls of both upper and lower layer at the distance of $2 \cdot 68$ Å. between oxygen atoms. This approach may be accounted for by hydrogen bonding between layers. Such interlayer contacts occur three times less than the contacts within the layer, which explains the perfect cleavage parallel to the layer plane. Moreover, the contacts between

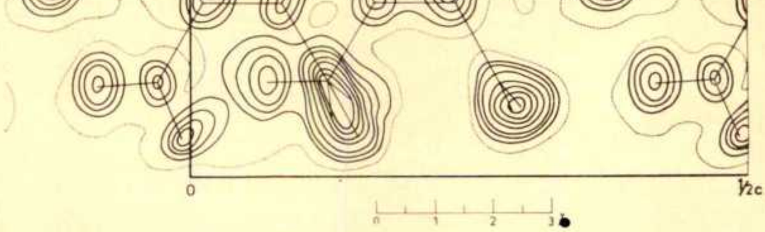

Fig. 1. The ($h0l$) electron-density projection of the crystal structure of mellitic acid. Contour interval, $2e.\text{Å}.^{-2}$; zero contour, dotted

carboxyls within the molecule itself occur at distances of 2·69, 2·71 and 2·75 Å. between oxygen atoms. Consequently, all oxygen atom contacts in the crystal structure are not essentially different, varying from 2·62 to 2·75 Å. If we assume, therefore, that the hydrogen bonding may be effectuated by all these contacts, then there are in the structure more hydrogen bonds than protons. This is possible only if a randomness of the proton positions is admitted. In favour of this assumption is also the symmetry of the carboxyls, since only one value of 1·23 ± 0·01 Å. for C–O distance has been found for all carboxyls and not only for those located on diad axis. The insignificant difference in length of the C–O bonds was previously established only for ionized carboxyls[4]. The symmetry of the carboxyl as well as a slightly shorter C–O bond found in the present case may be explained by the unique molecular structure of mellitic acid, which enables all carboxyls to be engaged in a hydrogen bond network throughout the crystal structure.

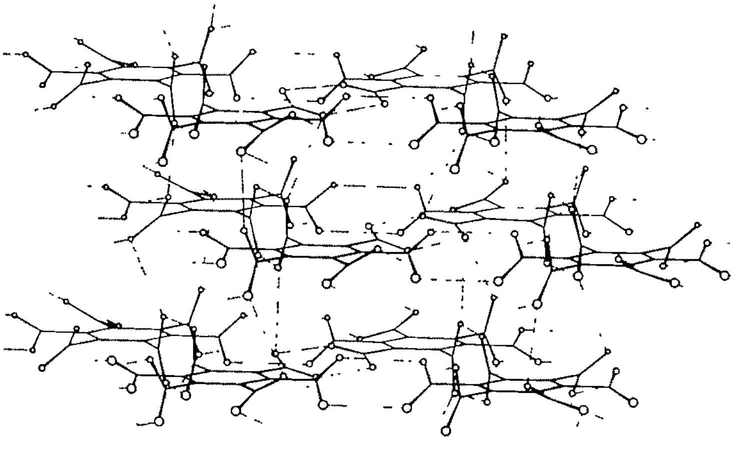

Fig 2. The layers of the crystal structure of mellitic acid as viewed in the direction of the *a*-axis. The separation between layers is 1/2*b*. The hydrogen bonds between oxygen atoms (circles) are shown by dotted lines

The more accurate determination of the bond-lengths has been prevented by overlapping of the atoms in the electron density projection. The three-dimensional Fourier synthesis as well as the difference synthesis is therefore anticipated. The C–C bond-lengths within the benzene ring as well as from it to carboxyl, obtained from the projections, are in good agreement with the previously reported data for this kind of compound, that is, 1·40 ± 0·01 Å., and 1·50 ± 0·01 Å., respectively. The reliability indices are: $R(h0l) = 0·12$; $R(0kl) \doteq R(hk0) = 0·13$.

We wish to thank Miss N. Katinčić for carrying out the synthesis of mellitic acid starting from xylene. We are also indebted to Mrs. I. Guštak-Mašek for the micro-analysis and to Mrs. G. Finžgar for valuable help in the computation.

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[1] Deverin, L., *Bull. Soc. Vaud. Sci. Nat.* (Lausanne), 59, 417 (1937).

[2] International Tables for X-Ray Crystallography, 1 (The Kynoch Press, Birmingham, 1952).

[3] Bezjak, A., *Acta Cryst.*, 12, 765 (1959).

[4] van Bommel, A. J., and Bijvoet, J. M., *Acta Cryst.*, 11, 61 (1958). Marsh, R. E., *ibid.*, 11, 654 (1958). Hahn, T., and Buerger, M. J., *Z. Krist.*, 108, 130, 419 (1957).

Thermal Expansion of Triglycine Sulphate

Matthias, Miller and Remeika[1] were the first to observe that triglycine sulphate becomes ferroelectric below 47° C. The dielectric properties and the specific heat of this crystal have been studied through the transition temperature by Hoshino, Mitsui, Jona and Pepinsky[2]. The observed variation of the dielectric properties as a function of temperature in this crystal shows that the transition is of second order. Hoshino *et al.* concluded that the anomaly is not of the λ-type, since their specific heat – temperature curve showed only a hump. It was decided to investigate the thermal expansion of this crystal as it might throw some light on the nature of the transition.

Triglycine sulphate belongs to the monoclinic system. Wood and Holden[3] determined the unit cell dimensions and the optical properties of this crystal. The complete structure of the ferroelectric phase of this crystal has been determined by Hoshino, Okaya and Pepinsky[4]. The crystallographic axial system of these authors is used in this communication in designating the directions in this crystal. The thermal expansion of triglycine sulphate and its temperature variation have now been investigated both above and below the transition point by the interferometric method for the following directions in the crystal: (1) parallel to the *b* axis, which is also the axis of spontaneous polarization in the crystal, and (2) parallel to the *a* and *c* axes, and perpendicular to the *a* axis in the (010) plane.

The specimens used for the expansion measurements along these four directions were cut from different crystals of triglycine sulphate and the measurements were repeated several times for each direction.

The variation of the thermal expansion coefficient (α) with temperature (T) for these four directions is plotted in Fig. 1. The results are summarized as follows:

Along the *c* and *a* axes and perpendicular to the *a* axis, the crystal expands as it is heated up to the transition point, the expansion coefficient becoming larger and larger as the transition point is approached. At the transition point there is an abrupt reversal of the sign of the expansion coefficients, the crystal contracting along these directions above the transition point. The behaviour along the *b* axis is just the opposite. The crystal contracts on heating up to the transition point and thereafter expands in this direction.

From the continuous change in the dimensions of the crystal, it appears that the transition is of the second order. This result is in agreement with that reported by Hoshino *et al.*[2]. From the nature of the α versus T curves (Fig. 1) one has to infer that the anomaly is of the λ-type, which does not agree with the observations of Hoshino *et al.*[2] on the specific heat.

Ubbelohde and Woodward[5] have found that in Rochelle salt the direction of maximum expansion coefficient lies almost along the direction of the shortest hydrogen bond. From the structural data of Hoshino, Okaya and Pepinsky[4] and the present

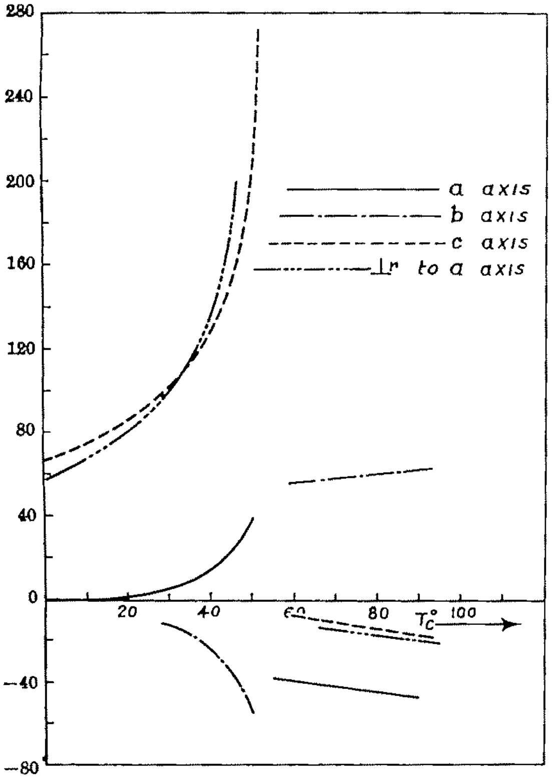

Fig. 1. α v. T curves for triglycine sulphate

results on the thermal expansion a similar conclusion could be drawn, namely, the direction of maximum expansion coefficient coincides nearly with the direction of the shortest hydrogen bond (length 2·438 Å.), which according to Hoshino *et al.*[4] is responsible for the dipole reversal mechanism.

The temperature at which the abrupt change in sign of α occurs has been observed to be slightly different for measurements along different directions, but all of them lie between 51° C. and 53° C., which is definitely higher than the value of the transition temperature of 47° C. reported by Matthias *et al.*[1] and Hoshino *et al.*[2]. It should be mentioned that all the measurements reported here were carried out while the crystals were heated. Taking measurements while the crystal was being cooled, it was found that the change in sign of α occurred at about 47° C. Preliminary evidence has also been obtained for thermal hysteresis in this crystal. Further investigations are under way to study the thermal hysteresis and a detailed report of the results will be published in due course.

I wish to express my thanks to Prof. R. S. Krishnan for his interest and to Dr. Vedam and Dr. Srinivasan for discussions. I also thank the former for suggesting the problem.

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[1] Matthias, B. T., Miller, C. E., and Remeika, J. P., *Phys. Rev.*, **104**, 849 (1956).

[2] Hoshino, S., Mitsui, T., Jona, F., and Pepinsky, R., *Phys. Rev.*, **107**, 1255 (1957).

[3] Wood, E. A., and Holden, A. N., *Acta Cryst.*, **10**, 145 (1957).

[4] Hoshino, S., Okaya, Y., and Pepinsky, R., *Phys. Rev.*, **115**, 323 (1959).

[5] Ubbelohde, A. R., and Woodward, I., *Proc. Roy. Soc.*, A, **185**, 448 (1946).

CHEMISTRY

Recrystallization of Amorphous Copper Sulphate Monohydrate

When copper sulphate pentahydrate is dehydrated in a vacuum at room temperature, the resulting product, of composition copper sulphate monohydrate, possesses no evident crystal structure as shown by X-ray examination[1]. When the amorphous product is heated, crystallization occurs as shown by the appearance of the X-ray lines of the crystalline monohydrate[2].

The changes which occur on heating can also be followed by investigating the changes in the electron spin resonance spectrum, since the spectrum of the paramagnetic copper ion is influenced by its environment in the solid which, of course, will change as crystallization occurs. The spectra were recorded on a 'Varian Model 4500' spectrometer. The curves in Fig. 1 are traces of the derivative of the absorption curve as recorded by this instrument.

Copper sulphate pentahydrate was ground to pass a 200-mesh sieve. Curve *F* illustrates the electron spin resonance spectrum on this powdered material. Curve *A* was obtained from a portion of this material which had been dehydrated in a vacuum desiccator over magnesium perchlorate until the composition corresponded to the monohydrate. The sample showed a very weak broad absorption. The extreme broadening was presumably due to the random environment of the paramagnetic copper ions in the amorphous material. When the material is heated, changes in the spectrum occur after an apparent induction period. Curves *B*, *C* and *D* indicate the

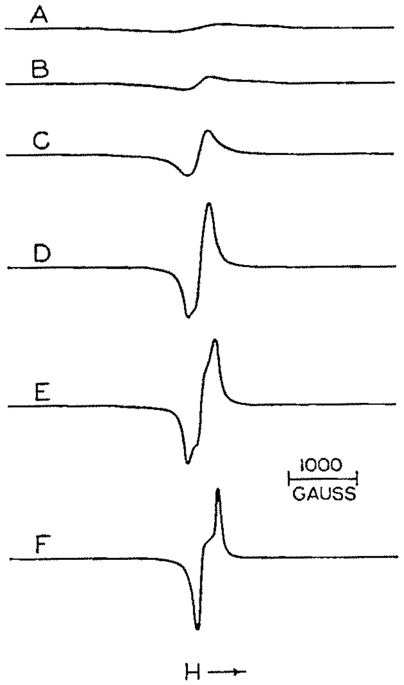

Fig. 1. Electron spin resonance spectra of copper sulphate hydrates. *A*, $CuSO_4.H_2O$ freshly prepared; *B*, $CuSO_4.H_2O$ heated 170 min. at 75° C.; *C*, $CuSO_4.H_2O$ heated 190 min. at 75° C.; *D*, $CuSO_4.H_2O$ heated 200 min. at 75° C.; *E*, Crystalline $CuSO_4.H_2O$ prepared at 110° C.; *F*, powdered $CuSO_4.5H_2O$.
Curves *A*, *B*, *C* and *D* were recorded at twice the amplification of curves *E* and *F*

traces obtained when the length of time of heat treatment of the sample at 75° C. was increased. With the recrystallization of the solid, the absorption narrows. Curve *E* is for a sample which was prepared and completely recrystallized at 110° C. A small amount of fine structure on the low-field side of the maximum is evident in curves *D* and *E*.

X-ray powder diagrams were taken at various stages of the recrystallization, using a General Electric recording X-ray diffraction unit. The material which gave the electron-spin resonance spectrum of curve *A* showed no diffraction lines. After heating, the X-ray diffraction lines of the monohydrate appeared.

It is interesting to note that the electron-spin resonance spectrum of a sample only partially recrystallized could not be simulated by the spectrum of a mixture of the amorphous monohydrate and the completely recrystallized compound. Even under higher amplification, the spectrum of sample *C* in Fig. 1 shows no evidence of fine structure on the low-field side of the maximum. A mixture of 19 per cent of the recrystallized monohydrate (sample *E*) and 81 per cent of the amorphous material (sample *A*), which gave the same overall intensity of absorption, very clearly showed fine structure. The very first stages of recrystallization do not appear to involve the formation of regions in the solid which yield the same electron-spin resonance spectrum as the completely recrystallized material of sample *E*.

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[1] Kohlschutter and Nitschmann, *Z. phys. Chem.*, Bodenstein Festband, 494 (1931).

[2] Frost, Moon and Tompkins, *Canad. J. Chem*, 29, 604 (1951).

Some Atomic Reactions by Absorption Spectroscopy

NORRISH and co-workers[1-3] have shown that vibrationally excited oxygen molecules are produced by flash photolysis of chlorine dioxide, nitrogen dioxide and ozone. These molecules are believed to originate in reactions of the type:

$$O + ClO_2 \rightarrow ClO + O_2^* \quad (1)$$

Combination of familiar atomic flame techniques[4,5] with absorption spectroscopy should enable such reactions to be studied directly. We have made observations of the reactions of atomic oxygen with ClO_2, NO_2 and O_3 in this way, at pressures of 0·1–30 mm. mercury. Similar studies have been made using atomic nitrogen and hydrogen in the cases of ClO_2 and O_3.

Atoms were produced from the molecular gas by electrodeless discharges (at 2,450 or 30 Mc./s.) and mixed with the reactant gas at one end of a reaction cell, 2 cm. in diameter and 30 cm. long, with quartz end-windows. Linear flow-rates through the cell were between 100 and 250 cm./s. The continuum from a high-pressure xenon source was focused through this cell, and viewed with a Hilger quartz, Littrow type, spectrograph. The extent to which atomic oxygen had reacted could be seen from the decrease in the green after-glow (caused by reaction of O with NO formed in the discharge from nitrogen impurities) when the reactant was added, and, in the case of atomic nitrogen, from the decrease in the Rayleigh after-glow. In most cases spectroscopic data were obtained at flow-rates such that these after-glows were almost extinguished, that is, at complete consumption of the atoms. Under these conditions weak flames were usually seen in the reaction vessel.

Some observations are summarized in Table 1.

Table 1. SPECTRAL OBSERVATIONS OF SOME ATOMIC FLAMES

Atom	Reactant		
	ClO_2	NO_2	O_3
O	ClO_2 removed ClO produced Red flame	NO produced	Red flame (weak)
N	ClO_2 removed ClO produced (weakly) Green flame	—	O_3 removed No NO Blue-white flame
H	ClO_2 removed No ClO or OH Green flame (weak)	Red flame	O_3 removed No OH Red flame (weak)

The sensitivity of the spectroscopic system can be judged from the fact that amounts of NO and NO_2 giving partial pressures of 0·01 and 0·10 mm. mercury respectively could be observed readily. The rotational temperature of the NO produced in the $NO_2 - O$ reaction was the same as the temperature of the reaction vessel. A tenfold increase in pressure by the addition of argon gave no new information. No evidence was found for the existence of vibrationally excited species.

A quantitative examination of the system is necessary to see what limitations might apply to the production and detection of vibrationally excited oxygen. By adding sufficient NO_2 to quench the after-glow in the products of the oxygen discharge[6], and making spectroscopic determinations of the NO molecules produced, the concentration of atomic oxygen was established to be 10 per cent of the gases from the discharge at 2 mm. mercury. Although the efficiency of O_2 in deactivating O_2^* is not known, one collision in 10^5 can be taken as reasonable in comparison with the efficiencies given by Norrish[3] for other molecules. If it is further assumed that all the O atoms form O_2^* by reaction (1) the total number of O_2^* molecules in the reaction vessel is calculated to be $3 \cdot 6 \times 10^{16}$ at 2 mm. mercury, or 0·6 per cent of all molecules present.

An estimate of the amount of the other product of reaction (1) can also be made in the case of ClO, for which radicals the rate constant for recombination has been determined[7]. It gives a total number of ClO radicals of $2 \cdot 7 \times 10^{16}$, or 0·4 per cent of the gas at 2 mm. mercury. Our failure to find O_2^*, while ClO was readily detected, may be due to: (*a*) a deactivation efficiency of O_2 greater than the estimated 1 in 10^5; (*b*) a smaller absorption coefficient for O_2^* than for ClO; or (*c*) O_2^* not being produced by reaction (1), which possibly requires metastable O atoms rather than the ground-state atoms given by our discharges. Similar reasoning can also be applied to the $NO_2 - O$ reaction, except that now the NO produced, since it is stable, reaches 10 per cent of the total gas.

Concentrations of atomic nitrogen were measured by titrating the active nitrogen with nitric oxide[8], and afterwards determining the amounts of NO by spectral absorption when the discharge was extinguished. At 20 mm. mercury about 1 per cent nitrogen atoms were produced. The disappearance of ClO_2 by reaction with N atoms, but only very weak ClO, and no NO absorption can be interpreted by a low rate of:

$$N + ClO_2 \rightarrow NO + ClO \qquad (2)$$

or by rapid subsequent disappearance of the products of this reaction. Lack of NO absorption in the reaction of N with O_3 can be explained as follows. When [N] is greater than $[O_3]$, the nitric oxide formed is destroyed rapidly by the excess of free atoms. On the other hand, when ozone is in excess it will react rapidly with NO (ref. 9).

The concentration of atomic hydrogen was not measured, but previous experience[10] with the same type of discharge indicates a concentration of about 10 per cent. Again, the decrease in ClO_2 absorption, without any OH or ClO absorption, can be taken as evidence that the initial attack does not produce these radicals or that their subsequent reactions are extremely rapid.

The ease of detection of small quantities of NO and ClO suggests that absorption spectroscopy could be a useful tool for studying kinetics in steady-state flow systems of this type. Moreover, the spectroscopic measurement of NO has proved to be very useful in determining the concentrations of oxygen and nitrogen atoms. The visible reaction flames suggest that a study of their emission spectra may provide information regarding the initial steps in these reactions.

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[1] Lipscomb, F. J., Norrish, R. G. W., and Thrush, B. A., *Proc. Roy. Soc.*, A, **233**, 455 (1956).

[2] McGrath, W. D., and Norrish, R. G. W., *Proc Roy. Soc.*, A, **242**, 265 (1957).

[3] Norrish, R. G. W., *Proc. Chem. Soc.*, 247 (1958).

[4] Gaydon, A. G., and Wolfhard, H. G., *Proc. Roy. Soc.*, A, **213**, 366 (1952).

[5] Ferguson, R. E., and Broida, H P., Fifth Symposium on Combustion, 754 (Reinhold Pub. Corp., New York, 1955).

[6] Elias, L., Ogryzlo, E. A., and Schiff, H. I., *Canad. J. Chem.*, **37**, 1680 (1959).

[7] Edgecombe, F. H. C., Norrish, R G. W., and Thrush, B. A., *Chem. Soc. Spec. Pub.*, 9, 121 (1958).

[8] Kaufman, F., *J. Chem. Phys.*, **28**, 992 (1958).

[9] Ford, H. W., Doyle, G. J., and Endow, N , *J Chem. Phys.*, **26**, 1336 (1957).

[10] Shaw, T. M., "Formation and Trapping of Free Radicals", Chapter 3 (Academic Press, New York, 1960).

Ionic Hydration in Mixtures of Strong Acids and Water

FROM Raman and nuclear magnetic resonance measurements the concentrations of species present in mixtures of strong acids with water have been evaluated[1-3].

These measurements show that each ionic constituent has a maximal concentration at a certain composition of the mixture. The existence of such maxima can be understood in terms of hydration of the ionic species present.

Consider the following reaction:

$$HA + n\,H_2O \rightleftarrows H^+(H_2O)_{n+} + A^-(H_2O)_{n-} \qquad (1)$$

$$n_+ + n_- = n$$

Application of the law of mass action to (1) gives:

$$a_{H^+(aq.)} \cdot a_{A^-(aq.)} = K_n \cdot a_{HA} \cdot a_{H_2O}{}^n \qquad (2)$$

where $a_{H^+(aq.)}$ and $a_{A^-(aq.)}$ denote the activities of $H^+(H_2O)_{n+}$ and $A^-(H_2O)_{n-}$, and n is the total number of water molecules bound to the ions H^+ and A^-. By differentiation of (2) and combination with the Gibbs–Duhem equation, it can be shown that the activity product of the hydrated ions will have a maximum at the concentration[4]:

$$X^s{}_{HA} = \frac{1}{1 + n} \qquad (3)$$

where $X^s{}_{HA}$ is the stoichiometric mole fraction of acid. The maximum will thus appear at the stoichiometric concentration corresponding to the assumed composition of the hydrated ions.

When considering concentrations of ionic constituents, the variation of their activity coefficients will be neglected. From the electroneutrality condition:

$$C_{H^+(aq.)} = C_{A^-(aq.)} \qquad (4)$$

and (2) it can be shown that, within the uncertainty of the variation of the ionic activity coefficients, condition (3) is retained for the location in the maximal concentration of A^-.

The maxima can be considered to arise from the competition of various hydration reactions with the formation of HA molecules. It can be stated that the weaker the acid the more will the maximal ionic concentration be displaced in the direction of pure water. In Table 1 a comparison is made between experiments and equation 3.

Table 1. COMPARISON OF MOLE FRACTION OF MAXIMAL IONIC CONCENTRATION WITH EQUATION 3

n	$X^s{}_{HA}$ (3)	$X^s{}_{HA}$ exp. NO_3^-	ClO_4^-	HSO_4^-
1	0·50		0·40–0·50 (ref. 2)	0·42 (ref. 3)
2	0·33			
3	0·25	0·26 (ref. 5)		
4	0·20	0·21 (ref. 2)		

As seen from Table 1, the maximum ionic concentration of NO_3^- appears in the range where there are 3–4 H_2O per H^+, NO_3^-, while maximal concentration of ClO_4^- and HSO_4^- are in the range where there is about 1 H_2O per H^+, A^-. That nitric acid behaves differently from the other two acids is due to its weaker dissociation. From the data of Redlich *et al.*[2] it is found that even at $X^s{}_{HNO_3} = 0·40$ there are more than four water molecules per mole of dissociated acid.

In the primary dissociation of H_2SO_4, the main hydrated ionic constituent is likely to be H_3O^+ (ref. 6). However, the displacement of the maximal ionic concentration from 0·50 to 0·42 implies that hydrated species other than H_3O^+ are also present. This further hydration can be taken into account. By assuming the formation of all possible hydrated ionic species, it can be shown that maximum concentration of the anion A^- will be located at $X^s{}_{HA} = 1/(1 + \bar{n}')$, where $\bar{n}'$ approximates the average number of water molecules bound to the ions H^+ and A^- (ref. 4). For $X^s{}_{H_2SO_4} = 0·42$ this gives $\bar{n}' = 1·40$. The same value for $\bar{n}'$ was obtained by an approximate method of finding $\bar{n}'$ reported elsewhere[6]. Using the values of $\bar{n}'$ given in ref. 6, it is also found that at $X^s{}_{H_2SO_4} = 0·42$ the free water practically disappears and becomes tied up as water of hydration. Now, Young *et al.*[3] have found that measurable amounts of sulphuric acid start to appear at this same concentration. It thus seems that molecules of sulphuric acid cannot appear until nearly all free water has been consumed.

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[1] Chedin, J., *J. Chim. Phys*, 49, 109 (1952).

[2] Hood, G. C., Redlich, O., and Reilly, C. A., *J. Chem. Phys.*, 22, 2067 (1954).

[3] Young, T. F., Maranville, L. F., and Smith, M. M., in Hamer, W. J., "The Structure of Electrolytic Solutions", 35 (John Wiley, New York, 1959).

[4] Hogfeldt, E., *Acta Chem. Scand.* (in the press).

[5] McKay, H. A. C., *Trans. Farad. Soc.*, 52, 1568 (1956) (quoting measurements by Young and Krawetz).

[6] Hogfeldt, E., *Acta Chem. Scand.* (in the press).

The Sodium Silicate – Hydrogen Peroxide System

THE chemistry of silicic acid in solution is rather ill-defined[1] owing to the extremely weak nature of this acid[2] and its tendency to polymerize[3,4]. As there are many instances where hydrogen peroxide effectively prevents anionic polymerization[5,6], an attempt was made to investigate the hydrolysis of silicate ions in the presence of hydrogen peroxide by means of accurate conductivity[7] and *p*H measurements. For comparison, the same technique was applied to an identical series of solutions in the absence of hydrogen peroxide. Fig. 1 shows the results of the investigation on solutions which were 0·01 *M* in sodium silicate and, in the case of the 'peroxidized' system, approximately 0·1 *M* in hydrogen peroxide; sulphuric acid was 0·498 *M*.

The *p*H curve of the system sodium silicate–water (full lines) shows two changes: a weak one on addition of 4·75 ml. sulphuric acid and a strong change on addition of 9·3 ml. acid. The former change may be due to the reaction of the dihydroxy-*ortho*silicate ion[1–3] with hydrogen ions:

$$(SiO_2(OH)_2)^{2-} + H_3O^+ = (SiO(OH)_3)^- + H_2O \quad (1)$$

The latter change denotes obviously the completion of the neutralization reaction:

$$1OH^- + H_3O^+ = 2H_2O \quad (2)$$

the hydroxyl ions being those hydrolytically produced by sodium silicate. Corresponding changes in the conductivity curve of the same system are brought out by the interpolated point of intersection[8] *B* (equation 1) and point *D* (equation 2) respectively. The strong *p*H change (equation 2) seems to mask the final decomposition of the trihydroxy-*ortho*silicate ion to silicic acid, although some indication of the latter reaction is borne out by the shape of the conductivity curve near point *D*.

A similar picture emerges from the investigation of the sodium silicate – hydrogen peroxide system (dotted lines), with the difference that as the *p*H of the initial solution lies below 10·75 (owing to the consumption of hydroxyl ions by hydrogen peroxide in alkaline solution) the points *A* and *B* are absent. A comparison of the specific conductivities in the two systems for solutions containing 6–9 ml. sulphuric acid reveals the rather significant finding of higher conductivity in the 'peroxidized' system (*p*H 8–10) which may be attributed to the lack of polymerization of the silicate ion in this system. The suggestion is therefore made that the study of the hydrolysis of the silicate ions is rendered less complex in the presence of hydrogen peroxide.

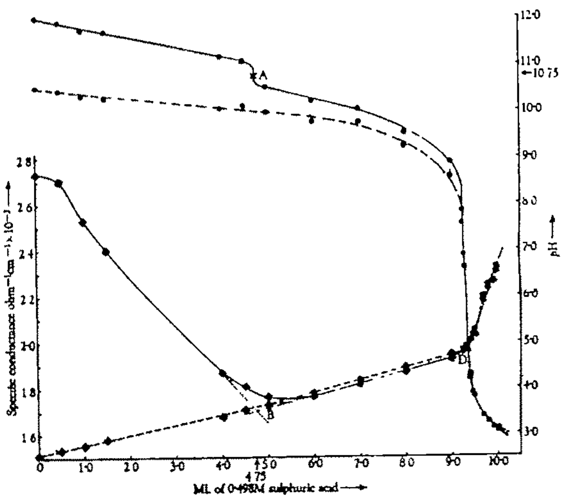

Fig. 1. Measurements of specific conductivity (◆) and *p*H (●) for the systems sodium silicate – hydrogen peroxide – water (dotted lines) and sodium silicate – water (full lines)

Detailed results of our investigation will be published elsewhere. We wish to acknowledge a research grant from Joseph Crosfield and Sons, Ltd., and the gift of pure unstabilized hydrogen peroxide from Laporte Chemicals, Ltd.

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[1] Greenberg, S. A. *J. Chem. Educ.*, 36, 218 (1959).

[2] Remy, H., "Treatise on Inorganic Chemistry", 1, 493 (Elsevier, Amsterdam, 1956).

[3] Thilo, E., *Angew. Chem.*, 63, 201 (1951).

[4] Alexander, G. B., *J. Amer. Chem. Soc.*, 76, 2094 (1954)

[5] Kakabadse, G., and Wilson, H. J., *Nature*, 180, 861 (1957)

[6] Jahr, K. F., "Fiat Review of German Science 1936–1946", Pt 3, 172.

[7] Constable, F. H., and Tegul, S., *Nature*, 160, 55 (1947).

[8] Kortum, G., and Bockris, J. O'M., "Textbook of Electrochemistry", 1, 216 (Elsevier, London, 1951).

Volatile Organic Matter in Algal Culture Media and Sea Water

DURING an investigation of the extracellular products of marine algae, in which filtered culture media were concentrated by vacuum distillation, the distillates, which had an appreciable odour, were found to have a greater absorption of ultra-violet light than distilled water. The absorption of distillates increased by distilling at atmospheric pressure with ice cooling, a spray trap being incorporated in the apparatus. Examples of spectra from filtrates of uni-algal cultures of *Enteromorpha* sp., *Phaeocystis pouchetii* and *Phaeodactylum tricornutum*, bacteria-free *Ectocarpus confervoides* and membrane-filtered sea water are shown in Fig. 1. The overall shape of the absorption curves is somewhat affected by rate of boiling, degree of cooling, alkalinity of sample and freshness of the distillate. This suggests that more than one constituent may be present.

By redistilling distillates under various conditions it is possible to estimate that the recovery of volatile ultra-violet absorbent material is about 50 per cent when a distillate of 250 ml. is collected from a sample of 2·5 l. Measurements with CH_3COOH, C_2H_5COOH,

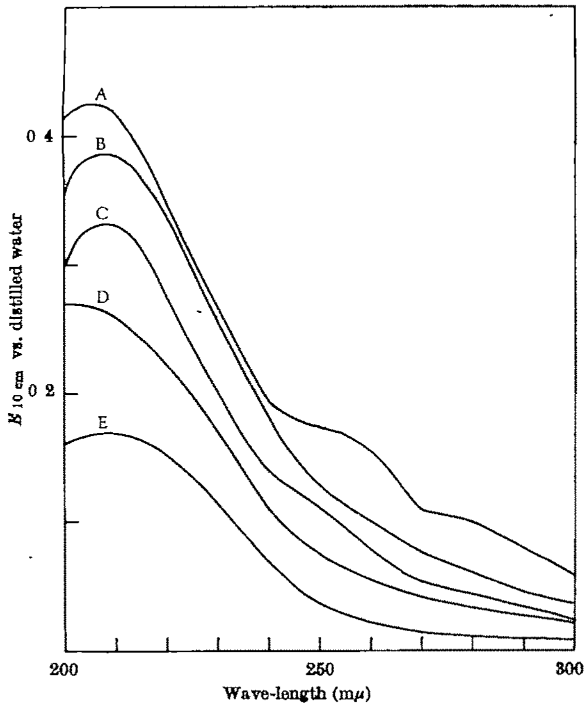

Fig. 1. *A*, *Ectocarpus* (× ½); *B*, *Phaeodactylum*; *C*, *Enteromorpha*; *D*, *Phaeocystis*; *E*, sea water

$CH_2OHCOOH$, H_3BO_3, $HCHO$, CH_3CHO, $(CH_3)_2CO$, $(CH_3)(C_2H_5)CO$, NH_3, CH_3NH_2, $(CH_3)_2NH$, $C_3H_7NH_2$ have shown that the absorptions found can only be accounted for if these substances are present (singly or as a mixture) at concentrations exceeding about 10 mgm./l. in the sample. Formic acid or trimethylamine at concentrations of about 1 mgm./l. could account for the ultra-violet absorption; but the former is unlikely to be volatile from sea water and the latter would necessitate larger carbon values than were actually found.

That the volatile material is at least partly organic is shown by carbon estimations carried out as follows. The distillates were boiled, after the addition of sodium hydroxide to retain carbon dioxide, and the vapours passed with a stream of oxygen through a simple 'empty tube' combustion train. The re-distillation with alkali causes the loss or retention in the flask of nearly 50 per cent of the ultra-violet absorptive material. This heavy loss suggests that in fact the carbon content of the volatile material may be a good deal higher than the figures in Table 1 indicate.

Table 1. CARBON AND SULPHUR DETERMINATIONS IN FILTERED CULTURE MEDIA AND IN SEA WATER

	Carbon (μgm./l.)	Sulphur (μgm./l.)
Ectocarpus confervoides		
(bacteria-free)		
Cultures I and II	—	36 and 33
Culture III	110	22
Enteromorpha sp.		
(uni-algal)	70	24
Phaeocystis pouchetii		
(uni-algal)	70, 100, 100	4·0, 6·0
Phaeodactylum tricornutum		
(uni-algal)		
Culture *A*	80	11, 9·5
Culture *B*	10, 50	2·5, 4·0, 3·0
Sea water, Lat. 50° 02′ N.,		
Long. 4° 22′ W.		
(English Channel)		
Sept. 8, 1959	—	1·0
Oct. 13, 1959	—	1·5
Nov. 11, 1959	40, 40, 50	2·5, 2·0, 3·0, 2·0
Dec. 1, 1959	30, 40, 20, 30	1·5, 2·5, 3·5, 4·0

The characteristically 'weedy' odour of the distillates suggested the presence of organic sulphides since Haas[1] has shown that these substances are released from some marine algae. Micro-estimations of sulphur were carried out as follows. The distillates were passed through a small column packed with sulphur-free sugar charcoal, which had been found to remove odour and ultra-violet absorbent material. The absorbed material was volatilized at 600° C. in a stream of hydrogen and passed through a tube furnace at 1,000° C. to convert sulphur compounds to hydrogen sulphide[2]. This was then determined colorimetrically by the 'Lauth's violet' method of Polson and Strickland[3]. Sulphur contents of 1–4 μgm. sulphur/l. were found in samples of offshore sea water and higher values in cultures (see Table 1). Hydrogen sulphide could not be detected in the distillates (less than 0·1 μgm. sulphur/l. in the samples). If the sulphur were present as dimethyl sulphide the quantities found are quite insufficient to account for all the ultra-violet absorption. Further, the carbon : sulphur ratio is greater than in this compound. The nature of the other carbon compounds which must be present is as yet unknown.

These volatile materials were present in the media from bacteria-free cultures of *Ectocarpus* and appear to be released by the alga. Also, since they can be detected early in the growth of cultures of *Ectocarpus*, *Phaeocystis* and *Phaeodactylum* they are probably produced during healthy growth.

Fogg and Boalch[4] have already shown the presence of considerable quantities of extracellular nitrogenous matter in *Ectocarpus* cultures, and have suggested that it may have importance as a chelating agent. If the volatile sulphur is present as dimethyl sulphide or related compounds (which are known to form complexes with heavy metal salts[5]), the metal-binding properties of the extracellular products may be enhanced and may partly account for the marked resistance of *Ectocarpus* to compositions containing copper and mercury[6].

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[1] Haas, P., *Biochem. J.*, 29, 1297 (1935).

[2] Fogo, J. K., and Popowsky, M., *Anal. Chem.*, 21, 734 (1949).

[3] Polson, D. S. C., and Strickland, J. D. H., *Anal. Chim. Acta*, 6, 452 (1952).

[4] Fogg, G. E., and Boalch, G. T., *Nature*, 181, 789 (1958).

[5] Challenger, F., "Aspects of the Organic Chemistry of Sulphur" (Butterworth, London, 1959).

[6] Harris, J. E., *J. Iron and Steel Inst.*, 148, 405 (1943).

Some Phenolic Constituents of Cigarette Smoke

PHENOL and a number of substituted phenols have strong tumour-promoting properties[1], and similar activity has recently been demonstrated in a phenolic fraction of cigarette smoke (Salaman, M. H., and Roe, F. J. C., private communication). It seemed of interest, therefore, to record some results we have obtained in an examination of phenols from the smoke of a variety of British cigarettes.

The cigarettes were smoked mechanically (2 sec. puff of 25 c.c. every 15 sec.) and the smoke was condensed in traps at −60°. Volatile material was removed from the condensate by co-distillation with methanol, and the remaining tar, in methanol, was cooled to 0° to precipitate waxes. The filtered solution, in ether, was extracted successively with dilute aqueous acetic acid, sodium bicarbonate and sodium hydroxide, and the material recovered from the sodium hydroxide extract was methylated with diazomethane. Carboxylic acid esters present in the crude methylation product were hydrolysed with boiling ethanolic potassium hydroxide and the phenol ethers distilled. The compounds listed in Table 1 were obtained from a fraction b.p. 150–300° (1·1 gm. from smoke of 5,000 cigarettes) by gas-liquid chromatography[2]. They were identified by preparation of crystalline derivatives which were compared with authentic specimens and, where possible, by infra-red and ultra-violet absorption measurements. The cresyl ethers were eluted together from the chromatogram; the free cresols, however, were easily separated by counter-current distribution between benzene and a phosphate buffer as described by Golumbic[3]. The methoxyacetophenones obtained from the chromatogram were contaminated with unidentified material but this was removed by crystallization of the 2 : 4-dinitrophenylhydrazones.

The cigarette smoke must have contained the parent phenols of the ethers listed in Table 1 (veratrole, of course, could have arisen from either catechol or guaiacol). Some of the phenols have already been detected in cigarette smoke by spectroscopic and other means[2,4,5], and the isolation of phenol, *m*-cresol and guaiacol as their 3 : 5-dinitrobenzoates from smoke of Japanese cigarettes was reported[5] after the completion of the present work. The hydroxyacetophenones, however, have not previously been found in cigarette smoke. It is of interest, with regard to the promoting activity of the phenolic fraction of cigarette smoke (Salaman, M. H., and Roe, F. J. C., private communication), that phenol itself and the three cresols have all been shown to have tumour-promoting properties.

Results closely similar to the above were obtained by Mr. G. W. J. Matthews in this Laboratory by gas-liquid chromatography of phenol ethers derived from smoke of a brand of Argentinian cigarette said to contain fermented tobacco.

Table 1. GAS-LIQUID CHROMATOGRAPHIC SEPARATION OF PHENOL ETHERS DERIVED FROM CIGARETTE SMOKE

Peak No.	Compound	Estimated mgm. in smoke from 5,000 cigarettes	Crystalline derivative prepared
1	Anisole	40	2 : 4-dibromoanisole, m.p. 60°
2	*o*-Cresyl methyl ether	100	*o*-cresyl-3 . 5-dinitrobenzoate, m.p. 137°
	m-Cresyl methyl ether		*m*-cresyl-α-naphthylurethane, m.p. 126°
	p-Cresyl methyl ether		*p*-cresylbenzoate, m.p. 70°
3	Veratrole	200	4-nitroveratrole, m.p. 95–96° 4 : 5-dinitroveratrole, m.p. 132°
4	Unidentified	120	
5	*m*-Methoxyacetophenone + unidentified	(10)*	2 : 4-dinitrophenylhydrazone, m.p. 189–190°
6	*p*-Methoxyacetophenone + unidentified	(5)*	2 : 4-dinitrophenylhydrazone, m.p. 229–230°

Column 6 ft. × 1 in., 20 per cent silicone grease *E* 301 on 80–100 mesh acid-washed 'Celite'; nitrogen flow-rate, 100 ml./min. Temperature was not measured, but was arranged so that anisole was eluted in about 10 min. Fractions were collected in traps at – 60° C.

* Estimated from weights of 2 : 4-dinitrophenylhydrazones.

Simple phenols have been found in cured tobacco leaf but they appear to be present in very small amounts[6]. The alkali-soluble fraction of an alcoholic extract of our cigarette tobacco afforded, after methylation, a significantly smaller amount of phenol ethers with boiling points below 300° C. than we obtained from the cigarette smoke, and it seems very likely that at least a proportion of the phenols in the smoke is produced by pyrolytic decomposition of other tobacco constituents.

We are indebted to Dr. J. W. Cook for his interest in this investigation.

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[1] Boutwell, R. K., and Bosch, D. K., *Cancer Res.*, 19, 413 (1959). Salaman, M. H., and Glendinning, O. M., *Brit. J. Cancer*, 11, 434 (1957).

[2] Carruthers, W., Johnstone, R. A. W., and Plimmer, J. R., *Chem. and Indust.*, 331 (1958).

[3] Golumbic, C., *J. Amer. Chem. Soc.*, 71, 2627 (1949).

[4] Rayburn, C. H., Harlan, W. R., and Hanmer, H. R., *Anal. Chem.*, 25, 1419 (1953). Commins, B. T., and Lindsey, A. J., *Brit. J. Cancer*, 10, 504 (1956). Bonnet, J., and Neukomm, S., *Helv. Chim. Acta*, 40, 717 (1957). Wynder, E. L., and Wright, G., *Cancer*, 10, 255 (1957). Clemo, G. R., *Tetrahedron*, 3, 168 (1958).

[5] Izawa, M., Kobashi, Y., and Sakaguchi, S., *Bull. Agric. Chem. Soc. Japan*, 23, 194 (1959).

[6] Onishi, I., and Yamamoto, K., *Bull. Agric. Chem. Soc. Japan*, 20, 61, 70 (1956); 21, 90, 181 (1957). Dymicky, M., and Stedman, R. L., *Tobacco Sci*, 3, 60 (1959).

BIOCHEMISTRY

Effect of Sulphydryl Group Reagents on Tryptophanase Activity

BOTH Gunsalus *et al.*[1] and Wada *et al.*[2] have reported that the presence of glutathione is advantageous during the assay of tryptophanase preparations. Wada *et al.* have also reported studies on the inhibition of tryptophanase by a number of SH-group reagents, and have suggested that the group is involved in the linkage between apoenzyme and substrate[3]. When we compare results which we have obtained at intervals since 1942, there is much agreement between us, but divergences, which may or may not be significant, do exist. Evans, Handley and Happold[4] reported a 12–20 per cent reduction of activity in washed cell preparation with sodium iodoacetate in a final concentration of $M/33$, and in later work with cell-free preparations activities were reduced by 20–25 per cent at concentrations of $5 \times 10^{-3} M$. At lower concentrations 5×10^{-5} and $10^{-6} M$ there has been no inactivation or even from time to time an apparent increase in the activity of the preparations. Further studies on inhibition were reported by Dawes and Happold[5]. Such preparations of tryptophanase are not necessarily free from other enzymes associated with pyridoxal phosphate-linked enzyme systems nor from the system which produced pyridoxal phosphate from pyridoxamine phosphate which Beechey and Happold described as a transaminase and which we now know contains a pyridoxamine phosphate oxidase similar to that found by Pogell in rabbit liver preparations; these divergences may be significant. During work in these laboratories on the purification of tryptophanase, the inhibition of activity by the SH-group alkylating reagent chloroacetophenone, and the reversal of SH-group inactivation, have been studied.

Table 1. Effect of Chloroacetophenone Concentration on Tryptophanase Activity

Chloroacetophenone concentration (*M*)	Tryptophanase activity	
	μgm. indole/5 min.	Percentage
0	25·5	100
$0·2 \times 10^{-6}$	25·5	100
$0·2 \times 10^{-5}$	24·3	95
$0·4 \times 10^{-5}$	14·3	56
$0·2 \times 10^{-4}$	6·9	27
$0·2 \times 10^{-3}$	0	0

The enzyme preparations used were obtained from *Escherichia coli*, and the tryptophanase activity was measured, as described by Beechey and Happold[6]. Reaction mixtures contained 0·5 ml. each of enzyme preparation (approximately 0·5 mgm.) protein; phosphate buffer (0·1 *M*, *p*H 7·8); potassium chloride solution (0·1 *M*); pyridoxal phosphate (100 μgm.); D,L-tryptophan ($1·96 \times 10^{-3}$ *M*, for L-form); and additions to 3·0 ml. Under these conditions at 37°, the rate of reaction was linear during the first 5 min., after which time 0·5 ml. of 10 per cent trichloracetic acid was used to stop the reaction. The indole produced was measured as previously described[6]. Pre-incubation with various reagents was carried out using reaction mixtures complete except for the addition of substrate.

Preliminary studies showed that chloroacetophenone at a final concentration of $0·2 \times 10^{-3}$ *M* required 20–30 min. pre-incubation for maximum inhibition. Using a 30-min. pre-incubation time, the effect of concentration of chloroacetophenone is shown in Table 1.

The effect of glutathione, at various concentrations, on the inhibition of tryptophanase activity by chloroacetophenone is shown in Table 2.

Table 2. Effect of Glutathione on Chloroacetophenone Inhibition of Tryptophanase

Glutathione concentration (*M*)	Tryptophanase activity (μgm. indole/5 min.), at various chloroacetophenone concentrations (*M*)				
	$0·2 \times 10^{-3}$	$0·4 \times 10^{-4}$	$0·2 \times 10^{-4}$	$0·4 \times 10^{-5}$	0
0	0·4	9·2	15·5	26·8	30·0
$0·2 \times 10^{-3}$	4·5	25·3	26·1	32·5	33·3
$0·2 \times 10^{-2}$	24·0	25·0	29·6	33·3	33·0

Glutathione was pre-incubated with enzyme for 60 min., after which chloroacetophenone was added and the mixture incubated for a further 30 min.

It can be seen that not only does glutathione protect tryptophanase from inhibition by chloroacetophenone, but also that it sometimes increases the activity of the preparation where no chloroacetophenone is present.

Further experiments showed that the extent of the increase in tryptophanase activity due to glutathione depended on the source and storage history of the preparation used. Maximum activation was found to occur at a glutathione concentration of $0·2 \times 10^{-2}$ *M*. When glutathione was replaced by cysteine in these experiments, initial results suggested progressive inhibition rather than activation. It was observed, however, that the yellow colour of reaction mixtures, due to pyridoxal phosphate, rapidly disappeared after the addition of cysteine. When pre-incubation of enzyme with cysteine was followed by the addition of co-factor only 5 min. before the addition of substrate, an increase in tryptophanase activity occurred. Here again, however, the extent of activation was variable, depending on the history of the enzyme preparation. Representative results are given in Table 3.

Table 3. Effect of Pre-incubation Time on the Activation of Tryptophanase by Cysteine

Pre-incubation time (min.)	Tryptophanase activity	
	μgm. indole/5 min.	Percentage of control
99·5	17·5	400
37·5	7·9	184
20·5	6·4	149
6·5	6·1	142
Control (no cysteine)	4·3	100

Pyridoxal phosphate was added at the end of pre-incubation, 5 min. before addition of substrate. Cysteine concentration was $0·2 \times 10^{-2}$ *M*.

It thus appears that cysteine enhances tryptophanase activity, presumably by reduction of oxidized SH-groups, but that this activation is masked by the removal of coenzyme, possibly by Schiff base formation. As the removal of coenzyme leads to the irreversible destruction of tryptophanase activity in the absence of metal ions, it appears that glutathione rather than cysteine should be used to protect the integrity of SH-groups during the purification of tryptophanase.

When sodium *p*-chloro mercuric-benzoate was used as an SH-group reagent inhibition showed itself at concentrations of 8×10^{-7} *M* and was almost absolute at 8×10^{-4} *M*. The tryptophanase activity was more than restored with 8×10^{-4} *M* glutathione.

Further work, which is continuing in these laboratories, indicates that cysteine protects tryptophanase from inhibition by chloroacetophenone. The reducing agent ascorbic acid has been found to inhibit tryptophanase activity under all conditions, but the reason for this remains obscure.

We wish to thank Miss Barbara Clough for technical assistance.

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[1] Gunsalus, I. C., Galeener, C. C., and Stamer, J. R., "Methods in Enzymology", 2, 238 (Acad. Press, 1955).

[2] Wada, H., Morisue, T., Sakamoto, Y., and Ichihara, K., *J. Vitaminol.* (Japan), 3, 183 (1957).

[3] Wada, H., *et al.*, Proc. Internat. Symp. on Enzym. Chem., Tokyo and Kyoto, 1957 (Pergamon Press, 1958).

[4] Evans, W. C., Handley, W. R. C., and Happold, F. C., *Biochem. J.*, 36, 311 (1942).

[5] Dawes, E. A., and Happold, F. C., *Biochem. J*, 44, 349 (1949).

[6] Beechey, R. B., and Happold, F. C., *Biochem. J.*, 66, 520 (1957).

Antigenicity of Œstrone–Protein Conjugates

The purpose of the present study was to determine whether steroids, when conjugated by stable covalent bonds to proteins, could act as haptenic groups in inducing steroid-specific antibodies. A detailed account of the method used for synthesizing œstrone–protein conjugates has been published elsewhere[1]. In brief, œstrone was converted to œstrone-17-*iso*cyanate and the latter was allowed to react with human and bovine serum albumin to yield the corresponding œstrone-17-carbamido-protein conjugates.

Rabbits were immunized with the œstrone–human serum albumin conjugate and the presence of precipitating antibodies in the sera was shown by the ring test. Two types of antibody were detected. One type was precipitated with œstrone–bovine serum albumin conjugate but not with bovine serum albumin and the reaction was inhibited by œstrone-17-carbamido-glycine. The other type was precipitated with human serum albumin but this reaction could not be inhibited with the glycine derivative of œstrone.

These results demonstrate that specific antibodies were produced to (1) the œstrone residue and (2) the protein carrier. The production of a third type of antibody, directed to the carbamido linkage itself, was ruled out since no reaction was obtained with butyl–carbamido–bovine serum albumin conjugate.

These conclusions were confirmed by three other methods.

(1) Dale–Schultz[2]: Guinea pigs were passively sensitized with rabbit antiserum produced to the œstrone-human serum albumin conjugate and their uteri were mounted in a Dale–Schultz apparatus. Addition to the bath of œstrone–bovine serum albumin conjugate and of human serum albumin resulted in the usual uterine contraction (Fig. 1*a*). However, addition of bovine serum albumin was without effect (Fig. 1*b*). Prior incubation of the uteri with the glycine derivative of œstrone completely inhibited the contraction due to the conjugate but not to human serum albumin (Fig. 1*c*).

(2) B.D.B.–Hæmagglutination: The œstrone–bovine serum albumin conjugate and human serum albumin were coupled to rabbit erythrocytes via *bis*-diazotized-benzidine[3,4]. The sensitized erythrocytes were added to serial dilutions of rabbit antiserum to œstrone–human serum albumin conjugate with and without various test inhibitors. Œstrone–bovine serum albumin conjugate as well as the glycine derivative of œstrone strongly inhibited hæmagglutination of erythrocytes sensitized with the conjugate while testosterone-17-glucoside and 17-hydroxycorticosterone-21-sodium succinate exhibited lower inhibiting activity. On the other hand, none of the test inhibitors mentioned had any effect on hæmagglutination of the erythrocytes sensitized with human serum albumin.

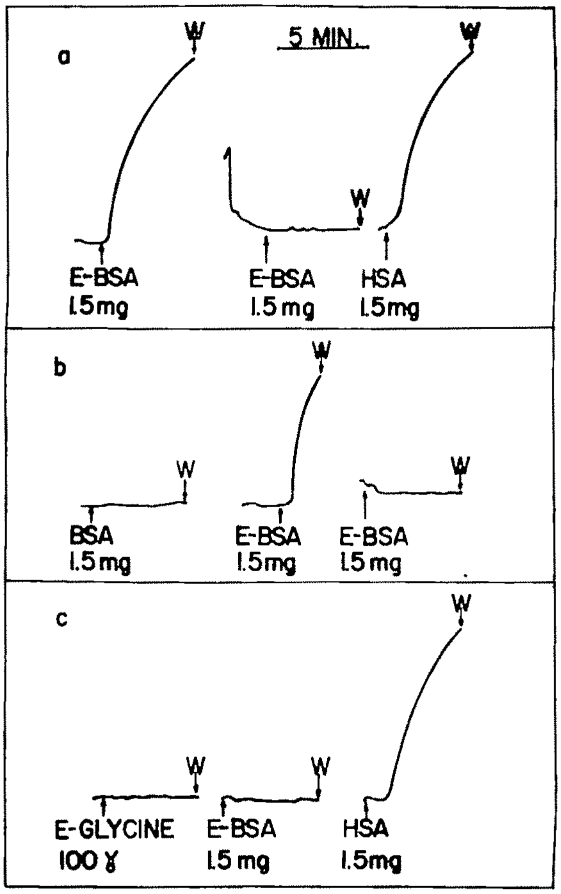

Fig. 1. *W*, Washing with Tyrode solution; *E-BSA*, œstrone–bovine serum albumin conjugate; *HSA*, human serum albumin; *BSA*, bovine serum albumin; *E-GLYCINE*, œstrone-17-carbamido–glycine

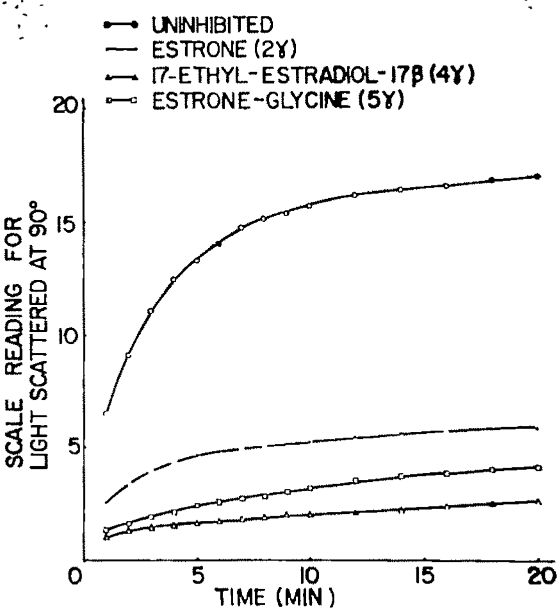

Fig. 2

(3) Turbidimetry: The course of the reaction between the conjugate of œstrone with bovine serum albumin and the rabbit antiserum to œstrone–human serum albumin conjugate was followed turbidimetrically by measuring light scattered at 90° with a Beckman *DU* spectrophotometer (cf. ref. 5). The curves obtained for the reaction at optimal antigen–antibody concentrations in the absence and in the presence of œstrone, the glycine derivative of œstrone and 17-ethynyl-œstradiol-17β are shown in Fig. 2. It can be seen that the steroids caused a marked reduction in turbidity; the percentage reduction in turbidity, due to these and other structurally related compounds, is given in Table 1.

The results presented demonstrate that the œstrone residue acts as a hapten when coupled to protein, and that the antibody site complementary to the œstrone residue is capable of accommodating not only œstrone but other structurally related molecules as well. These findings confirm in essence the conclusions of Beiser, Erlanger, Agate and Lieberman[6],

Table 1

Inhibitor	Amount added (γ)	Inhibition (per cent)*
Œstrone	2	66
Œstrone-glycine	5	85
Œstrone-glycine	10	99
17-Ethynyl-œstradiol-17β	4	76
17-Ethynyl-œstradiol-17β	8	89
17-Hydroxycorticosterone-21-sodium-succinate	200	31
Testosterone-17-glucoside	200	76
2,6-Dimethyl-phenol	200	0
3,4-Dimethyl-phenol	200	17

* Calculated on the basis of the turbidity readings at the 20-min. mark for the uninhibited and inhibited reactions (see Fig. 2).

who recently reported that they were able to produce specific antibodies to a series of steroid residues coupled to proteins.

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[1] Goodfriend, L., and Sehon, A. H., *Canad. J. Biochem. and Physiol.*, 36, 1177 (1958).

[2] Coulson, E. J., *J. Allergy*, 24, 458 (1953).

[3] Pressman, D., Campbell, D. H., and Pauling, L., *J. Immunol.*, 44, 101 (1942).

[4] Gordon, J., Rose, B., and Sehon, A. H., *J. Exp. Med.*, 108, 37 (1958).

[5] Boyden, A., Bolten, E. T., and Gemeroy, D. G., *J. Immunol.*, 57, 211 (1947).

[6] Beiser, S. M., Erlanger, B. F., Agate, jun., F. J., and Lieberman, S., *Science*, 129, 564 (1959).

Lack of Correlation between Synthesis of Nucleic Acids and Proteins in Denervated Muscle

After direct electrical stimulation of a muscle, non-collagen protein decreases and is later re-synthesized[1]. The rate of resynthesis is considerable, 1·9 mgm. of non-collagen protein/gm. of muscle tissue being resynthesized during 1 hr. The course of this accentuation of proteosynthesis was followed in the normal and denervated muscle. Fig. 1 shows that the decrease of proteins after stimulation of the muscle is the same in normal and denervated muscle and is accompanied by a decrease of ribonucleic acid, amounting to 15 per cent in the normal and 50 per cent in the denervated muscle. Such a decrease of ribonucleic acid and also an increase in proteolysis has been described after stimulation of nerve and brain tissue[2].

M. tib. anticus in rats was directly stimulated at a frequency of 300/min. for 6 min. Immediately, 2, 4 and 8 hr. after stimulation the muscles of the stimulated and control side were perfused with Tyrode solution. Following perfusion, both muscles were excised and the absolute amount of non-collagen proteins was determined (biuret method). The amount of ribonucleic acid was determined (Schneider's method) in another series of animals. The results are expressed in Fig. 1 as differences between the stimulated and control side. The first point of the curve corresponds to the difference between the muscle of the right and left side in control animals. Zero on the abscissæ represents the values immediately after stimulation. Proteins are expressed as μgm. non-collagen nitrogen/muscle. Ribonucleic acid values are expressed as μgm. ribonucleic acid phosphorus/100 mgm. protein nitrogen.

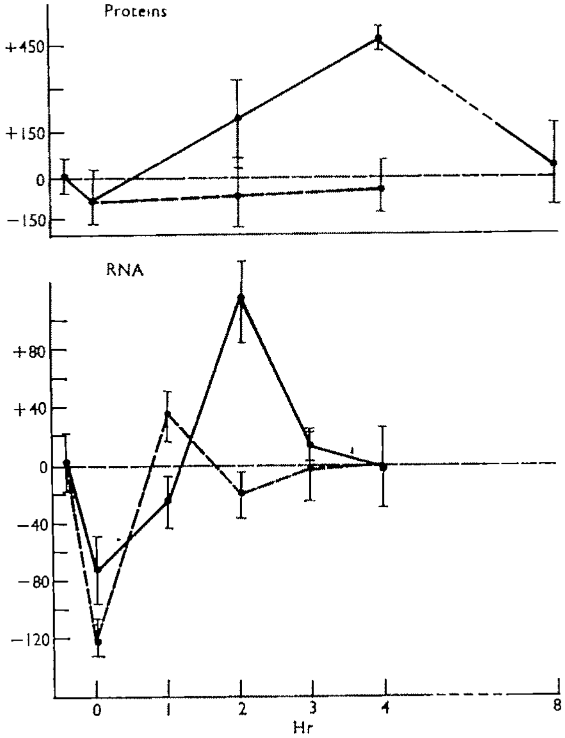

Fig. 1. Changes of non-collagen proteins and ribonucleic acid after stimulation of the normal (full line) and denervated (interrupted line) muscle

Fig. 1 shows that resynthesis of ribonucleic acid and proteins occurs after their breakdown in the normal muscle. However, the content of proteins and nucleic acids not only reaches the initial values but also their synthesis continues, the content of the proteins being 5 per cent above the initial values 4 hr. after stimulation, the content of ribonucleic acid being 22 per cent above the initial values 2 hr. after stimulation.

During the further course of the recovery phase following stimulation of the muscle, the content of proteins and ribonucleic acid returns to initial levels. The 'overshoot' reactions are apparently of general biological significance, since they have also been found during the process of resynthesis of glycogen[3] and during the restitution of the composition of ions in the muscle following stimulation.

These experiments also show that the recovery of ribonucleic acid following stimulation of the muscle precedes the resynthesis of proteins. Such a time correlation between the synthesis of ribonucleic acid and proteins has also been found in, for example, regenerating liver[5] and after the administration of growth-hormone[6]. After stimulation of the denervated muscle, resynthesis of ribonucleic acid, but not of the proteins, occurs.

There is therefore a certain lack of correlation between the resynthesis of proteins and ribonucleic acid in denervated muscle. The same lack of correlation was also observed during re-alimentation following starvation, which lowers both proteins and ribonucleic acid. Table 1 shows that the ribonucleic acid content is increased by 36·5 per cent within 12 hr. after re-alimentation and that resynthesis continues so that 60 hr. later ribonucleic acid increases by 80 per cent. Compared with ribonucleic acid, proteosynthesis was found to be delayed.

Table 1. CHANGES IN THE NET AMOUNT OF NON-COLLAGEN PROTEINS AND RIBONUCLEIC ACID IN M. TIB. ANTICUS OF RATS FOLLOWING A HUNGER PERIOD OF 10 DAYS AND AFTER RE-ALIMENTATION

Type of experiment	Proteins (µgm. non-collagen protein N/muscle)		Ribonucleic acid (µgm. ribonucleic acid P/muscle)	
	Right side	Left side	Right side	Left side
Control animals (6 rats)	9·2±0·4	9·0 ±0·5	41·6±0·6	41·0±0·4
	$P > 0·1$		$P > 0·1$	
	Before re-alimentation	After re-alimentation	Before re-alimentation	After re-alimentation
10 days hunger and 12 hr. re-alimentation (9 rats)	5·8±0·3	5·9 ±0·2	22·1±1·9	30·2±1·5
	$P > 0·1$		$P < 0·01$	
10 days hunger and 72 hr. re-alimentation (8 rats)	6·6±0·3	8·0 ± 0·2	20·6±0·7	37·0±1·4
	$P < 0·02$		$P < 0·001$	
10 days hunger and 72 hr. re-alimentation after denervation (16 rats)	6·7±0·2	6·3 ±0·3	22·8±1·4	29·4±1·0
	$P > 0·1$		$P > 0·001$	

In all animals the muscle of one side was removed at the end of the hunger period; the muscle of the contra-lateral extremity was removed 12 and 72 hr. after re-alimentation. Denervation was performed immediately before re-alimentation.

A significant increase in non-collagen protein was found only 72 hr. after re-alimentation. If the muscle is denervated immediately before re-alimentation, there is only an increase of ribonucleic acid but no increase of non-collagen protein after 72 hr. Similar results are thus obtained in both types of experiments.

The disturbance of proteosynthesis in denervated muscle could be due to the dissociation of the synthesis of nucleic acids and proteins.

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[1] Žák, R., Gutmann, E., and Vrbová, G., *Experientia*, 8, 80 (1957); *Physiol. bohemoslov.*, 6, 309 (1957).

[2] (*a*) Causey, G., and Stramann, C. J., *Biochem. J.*, 64, 29 (1956). (*b*) Ungar, G., Aschheim, E., Psychoyos, S., and Romano, D. V., *J. Gen. Physiol.*, 40, 635 (1957).

[3] Gutmann, E., Vodička, Z., and Vrbová, G., *Physiol. bohemoslov.*, 3, 182 (1954).

[4] Drahota, Z., Symposium on Nervous Regulation of Metabolism and Active Transport of Ions, Prague (1958)

[5] Eliasson, N. A., Hammarsten, E., Reichard, P., Aquist, S., Thorell, A. B., and Ehrensvärd, G., *Acta Scand. Chim.*, 5, 431 (1951).

[6] Cater, D. B., Holmes, B. E., and Mee, L. K., *Biochem. J.*, 66, 482 (1956).

Some Interrelated Aspects of Proline and Hydroxyproline Metabolism

PROLINE is known to be the sole source of hydroxyproline residues in collagen[1] and is converted to hydroxyproline only after its incorporation into a polypeptide[2,3]. If proline labelled with carbon-14 is employed as a specific precursor of hydroxyproline residues of collagen, the isotope concentration in urinary excretion products derived from such residues should reflect the rate of collagen catabolism. Pyrrole-2-carboxylic acid, an important product of hydroxyproline catabolism[4], as well as 'free' hydroxyproline and 'bound' hydroxyproline, was studied in these experiments. The present communication deals with the metabolic interrelationships between these urinary excretion products. For purposes of comparison the metabolic behaviour of proline and hydroxyproline residues of skin collagen is also considered.

The pertinent results are shown in Table 1. Some points are noteworthy: (1) The rate of turn-over of 'free' hydroxyproline is identical with that of pyrrole-2-carboxylic acid, thus extending the findings of Radhakrishnan and Meister[4]. (2) The rate showing the decrease of carbon-14 activity with time indicates that in the pools of 'free' hydroxyproline and pyrrole-2-carboxylic acid two metabolically distinct components are present which have a rapid and a slow turn-over rate, respectively. (3) During the days immediately following the injection of proline-U-^{14}C, the carbon-14-activity of the 'total' hydroxyproline fraction in urine is considerably lower than that of the 'free' hydroxyproline fraction. Thereafter the turn-over rate of the 'total' hydroxyproline fraction resembles that of the 'free' hydroxyproline and pyrrole-2-carboxylic acid fractions characterized by a slow turn-over rate. (4) The hydroxyproline residues in skin collagen are labelled rapidly. One day after injection of proline-U-^{14}C about 70 per cent of all proline residues destined to be hydroxylated have been converted to hydroxyproline. These findings are in agreement with those of Stetten and Schoenheimer[1] and are similar to those of Sinex and Van Slyke[5], who studied the conversion of lysine to hydroxylysine in collagen.

The existence of two metabolically distinct components in the 'free' hydroxyproline and pyrrole-2-carboxylic acid pools may be explained in at least two ways: (1) Two different metabolic pools of collagen may exist, one representing a labile fraction which is catabolized readily to pyrrole-2-carboxylic acid or 'free' hydroxyproline and the other constituting the source of 'bound' hydroxyproline. It is conceivable that the component having a rapid turn-over represents a transitory state preceding the stabile state of the collagen molecule. On the basis of the results presented, it might be speculated that proline residues are hydroxylated most rapidly when the polypeptide is in the labile, transitory state. (2) Because of its fibrous structure, collagen may not behave in accordance with the classical concepts of metabolic turn-over. Instead of the generally accepted mechanism for *de novo* synthesis of a macromolecule, the formation of new peptide bonds might be limited to the terminal sequences of amino-acid residues of the collagen fibres. This postulate is compatible with the experimentally observed non-uniform labelling of collagen by glycine-2-^{14}C [6] and phenylalanine-3-^{14}C [7]. It might, therefore, be postulated that the probability of metabolic activation of an amino-acid residue of collagen is dependent upon the distance at which the residue is located with respect to the metabolically active end of the fibre.

Table 1. SPECIFIC ACTIVITIES* OF URINARY EXCRETION PRODUCTS OF HYDROXYPROLINE RESIDUES AS WELL AS OF PROLINE AND HYDROXYPROLINE ISOLATED FROM SKIN COLLAGEN

	Days after injection of proline-U-^{14}C			
	1	2	3	6
Pyrrole-2-carboxylic acid	126	32	7·5	2·8
'Free' hydroxyproline	110	25	6·8	3·0
'Total' hydroxyproline	6·8	4·7	3·7	2·0
'Bound' hydroxyproline†		2·8	3·2	1·8
Proline of skin collagen	39		34·5	24·5
Hydroxyproline of skin collagen	22		24	21
Ratio of proline to hydroxyproline	0·56		0·7	0·88

Each of eight rats weighing 130 ± 4 gm. was injected with approximately 6·2 µc. of proline-U-^{14}C (specific activity, 7 µc./µM). Urine was then collected over several consecutive 24-hr. periods. The method devised for the isolation and determination of the urinary metabolites studied will be described in detail. Purification of skin collagen was carried out according to the procedures of Fitch (ref. 8).

* The specific activities are given as c.p.m./µM for 1 µc. of proline injected per 100 gm. animal weight.

† The specific activities of 'bound' hydroxyproline were computed from the values of 'total' and 'free' hydroxyproline using average values for the ratio of 'free' to 'total' hydroxyproline concentration (that is, approximately 1:13 in the urine of normally fed rats).

The results of these and other work on the metabolic behaviour of collagen will be published in detail elsewhere.

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[1] Stetten, M. R., and Schoenheimer, R. J., *J. Biol. Chem.*, 153, 113 (1944).

[2] Gould, B. S., and Woessner, J. F., *J. Biol. Chem.*, 226, 289 (1957).

[3] Gould, B. S., *J. Biol. Chem.*, 232, 637 (1958).

[4] Radhakrishnan, A. A., and Meister, A., *J. Biol. Chem.*, 226, 559. (1957).

[5] Sinex, F. M., and Van Slyke, D. D., *J. Biol. Chem.*, 232, 797 (1958).

[6] Gehrmann, G., Lauenstein, K., and Altman, K. I., *Arch. Biochem Biophys.*, 62, 509 (1956).

[7] Gehrmann, G., Lauenstein, K., and Altman, K. I., *Arch. Biochem. Biophys.* (in the press).

[8] Fitch, S. M., *Nature*, 176, 173 (1955).

Copper(II) Complexes of Histamine and 3-Methyl Histamine

In previous communications from this laboratory[1–3] evidence has been presented that acid dissociations occur from the 1 : 1 cupric complexes of histidine and histamine. In the presence of equimolar cupric chloride, one equivalent of alkali additional to that needed by either of the ligands alone is required to titrate all the acid produced. When the ligand/copper(II) ratio is 2 : 1 the amount of alkali required is the same as in the absence of the metal. From these facts it was concluded[2] that a co-ordinated water molecule and not the hydrogen attached to the unco-ordinated imidazole nitrogen was the source of the acid ionization from the 1 : 1 complexes. As a further test of this idea, we have investigated the copper(II) complexes of 3-methyl histamine.

Titration curves of 3-methyl histamine dihydrochloride in the absence and presence of cupric chloride are shown in Fig. 1. It can be seen that an acid ionization occurs from the 1 : 1 and not the 2 : 1 complex. Thus histamine and its 3-methyl derivative are similar in their co-ordinating properties with copper(II), and the interactions which occur may be represented thus:

$$\mathrm{Cu}^{2+} + L \underset{}{\overset{K_1}{\rightleftharpoons}} \mathrm{Cu}L^{2+} \qquad (1)$$

$$\mathrm{Cu}L^{2+} \overset{K_c}{\rightleftharpoons} \mathrm{Cu}L^{+} + \mathrm{H}^{+} \qquad (2)$$

$$\mathrm{Cu}L^{2+} + L \overset{K_2}{\rightleftharpoons} \mathrm{Cu}L_2{}^{2+} \qquad (3)$$

where

$$K_1 = \frac{[\mathrm{Cu}L^{2+}]}{[\mathrm{Cu}^{2+}][L]},\ K_c = \frac{[\mathrm{Cu}L^{+}][\mathrm{H}^{+}]}{[\mathrm{Cu}L^{2+}]},\ K_2 = \frac{[\mathrm{Cu}L_2{}^{2+}]}{[\mathrm{Cu}L^{2+}][L]}$$

The constants were calculated by the methods previously described[3] and the numerical values obtained did not vary significantly with pH (at 25° C. $pK_{a1} = 5{\cdot}80$, $pK_{a2} = 9{\cdot}90$, $\log K_1 = 9{\cdot}58$, $\log K_2 = 6{\cdot}56$, $pK_c = 7{\cdot}10$, where K_{a1} and K_{a2} are the acid dissociation constants of the ligand). These values are not very different from those reported for histamine[3].

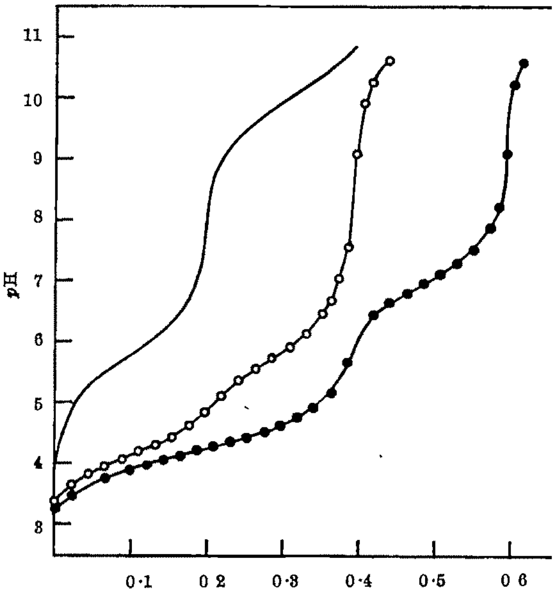

Fig. 1. Titration curves of 3-methyl histamine dihydrochlorid (0·2 mM in 20 ml.) with sodium hydroxide (0·11 N): ——, n additions; —○—○—○—, plus 0·1 mM cupric chloride —●—●—●—●—, plus 0·2 mM cupric chloride

The hydrogen attached to the unco-ordinat imidazole nitrogen cannot be the source of the ac dissociation from the histamine complex, since t hydrogen is not present in 3-methyl histamine and is most unlikely that the two ligands differ in t nature of the complexes they form. The eviden therefore, favours the previous suggestion that t acid dissociation (equation 2) occurs from a wat molecule co-ordinated to the 1 : 1 complex, CuL or the equivalent process, the uptake by this compl of a hydroxyl ion.

We are indebted to Dr. R. W. Schayer, Mer Institute of Therapeutic Research, for kindly suppl ing the 3-methyl histamine dihydrochloride used this investigation.

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[1] Datta, S. P., Leberman, R., and Rabin, B. R., *Biochem. J.*, 22*P* (1958).

[2] Leberman, R., and Rabin, B. R., *Nature*, 183, 746 (1959).

[3] Leberman, R., and Rabin, B. R., *Trans. Farad. Soc.*, 55, 1660 (195

ANIMAL PHYSIOLOGY

Renal Affection resulting from Blood Trauma in Extracorporeal Circuits

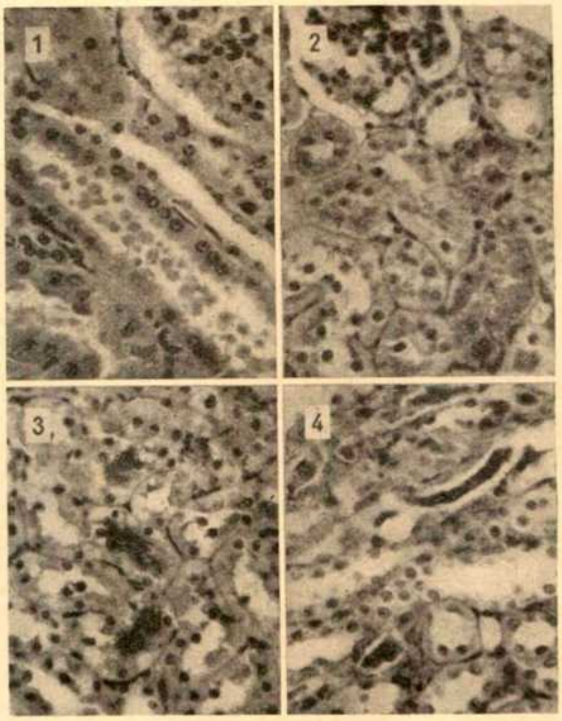
Previously, we have presented evidence that as result of the corpuscular blood trauma inherent extracorporeal circulatory systems including t heart–lung machines, adenosine triphosphate a 5-hydroxytryptamine (serotonin) are released in plasma, the former being derived mainly from t hæmolysed red cells and the latter from the disi tegrated platelets. Concurrently, the urinary exc tion of 5-hydroxytryptamine was increased, wh rather unexpectedly, there was a reduction in t excretion of 5-hydroxyindoleacetic acid, the main excretory product of hydroxytryptamine[1]. Since intravasal hæmolysis caused in any way may evoke an acute tubular ('lower nephron') nephrosis[2,3] and since, on the other hand, 5-hydroxyindoleacetic acid has been reported to be excreted also by active tubular secretion[4], we have studied the renal changes in a part of the animals, in which the above blood and urinary changes were found.

The animal material studied comprised four rabbits and three dogs. The experimental procedure was described in our previous report[1]. In brief, the animals were pretreated with pethidine hydrochloride, anæsthetized with pentobarbitone sodium and heparinized, following which an arteriovenous (femoral artery-superficial jugular vein) shunt, consisting of a specially prepared polythene tube, was created. In the period of this extracorporeal circulation, maintained for 90 min., the blood and urinary changes were observed. In general, the animals were killed at the termination of extracorporeal circulation, except one dog, in which the experiment was repeated one week later. At autopsy, both the kidneys were excised and fixed in neutral 10 per cent formalin. The embedded material was serially sectioned and stained with hæmalum–eosin and hæmalum–periodic acid Schiff-orange *G*. Frozen sections were stained with a mixture of sudan III and sudan IV for fats and with benzidine for hæmoglobin.

Pathological renal changes were found in both rabbits and dogs. In rabbits the changes generally consisted of some glomerular congestion associated with a leakage of eosinophilic, periodic acid–Schiff-positive material into the glomerular space (Fig. 1). The proximal convoluted tubuli showed a more or less intense intracellular deposition of orange *G*-positive material (Fig. 2), which also gave a positive benzidine-reaction. In addition, eosinophilic, orange *G*-positive material, in the form of granular bodies, droplets (Fig. 1) or plugs (Fig. 3), were seen especially in the lumina of the proximal convoluted tubuli. In the lower portions of tubuli there were eosinophilic, intensely periodic acid–Schiff-positive casts (Fig. 4). No definite destructive changes were observed in the tubular epithelium ; but in the intertubular vascular bed of the medulla there was a patchy type of congestion with occasional thrombosis. Fatty changes were consistently lacking. In dogs, changes of the same general type were encountered ; but they were less pronounced and the patchy vascular changes were absent. In the three rabbits and three dogs studied for comparison none of the changes described were seen.

Accordingly, the pathological renal changes observed largely correspond to those typical of the so-called 'lower nephron nephrosis'[2,3]. The lack of any definite destructive tubular changes may well be related to the short reaction time in the present experiments. The observation that the renal changes were less pronounced in dogs agrees with our finding, previously reported[1], that the corpuscular and humoral indices of blood trauma also were less pronounced in dogs. This also demonstrates that the renal changes under consideration are pathogenetically related to the blood trauma observed in our animals[1].

The glomerular and tubular structures were not affected throughout. On the other hand, the dog, in which the experiment was repeated one week later, did not show any pathological renal changes suggestive of irreversibility. Thus, the renal changes described can scarcely be regarded as being serious, especially *quo ad vitam*. Yet the present finding may be of significance at least in two respects. First, the unexpected reduction in the excretion of 5-hydroxyindoleacetic acid, as observed in our animals[1], may be related to some injury to the proximal convoluted tubuli, to which monoamine oxidase, essential for the conversion of 5-hydroxytryptamine into 5-hydroxyindoleacetic acid, is localized in the kidney[5] and in which the active secretion of 5-hydroxyindoleacetic acid presumably takes place[4]. In this respect, however, definite conclusions are not warranted, because the plasma-levels of 5-hydroxyindoleacetic acid were not observed in our animals and because detailed information on the mode of excretion of 5-hydroxytryptamine is not available for the present. Secondly, as pointed out previously[1], the corpuscular blood trauma in the present experiments was of the same general degree as that in the current heart–lung machines. A comparable degree of renal damage may therefore occur following the use of the current heart–lung machines for open cardiac surgery. Such renal damage may even be aggravated, should severe hypotension, as has been reported[6], ensue as a post-perfusion complication. It may be recalled that Daly and associates[7] found more severe renal changes, with occasional tubular necroses, in dogs perfused for about 3 hr. by means of a pump-oxygenator. They further found congestion, hæmorrhages and necrobiotic changes in other vital organs and were inclined to ascribe these changes as well as the allied signs of peripheral circulatory shock to 'vasotonins' appearing in the blood during perfusion. Our findings, previously discussed[1], suggest that adenosine triphosphate and 5-hydroxytryptamine may be the 'vasotonins' involved.



Figs. 1–4

Finally, Brull and Louis-Bar[8] have recently established the toxicity of artificially circulated, heparinized blood to the kidney and speculated that

5-hydroxytryptamine released from disintegrated platelets might be the causative agent. In their experience, furthermore, this toxic action could not be prevented by siliconizing the foreign channels.

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[1] Sarajas, H. S. S., Kristoffersson, R., and Frick, M. H., *Nature*, **184**, 1127 (1959); *Amer. J. Physiol.*, **197**, 1195 (1959).

[2] Burch, G. E., and Ray, C. T., *Ann. Int. Med.*, **31**, 750 (1959).

[3] Ogilvie, R. F., "Pathological Histology" (E. and S. Livingstone, Ltd., Edinburgh, 1951).

[4] Despopoulos, A., and Weissbach, H., *Amer. J. Physiol.*, **189**, 548 (1957).

[5] Blaschko, H., and Hellmann, K., *J. Physiol.*, **122**, 419 (1953).

[6] Schmutzer, K. J., Marable, S. A., Ruschke, E., Maloney, J. V., and Longmire, W. P., *Langenbecks Arch. klin. Chir.*, **290**, 64 (1958).

[7] Daly, I. De Burgh, Eggleton, P., Hebb, Catherine, Linzell, J. L., and Trowell, O. A., *Quart. J. Exp. Physiol.*, **39**, 29 (1954).

[8] Brull, L., and Louis-Bar, D., *Arch. Internat. Physiol. et Biochem.*, **65**, 470 (1957).

Loss of Heat by Evaporation in Young Lambs

Animals exposed to temperatures approaching or exceeding that of the body can lose little or no heat by radiation, convection and conduction; they must rely partly or wholly on evaporative cooling from the skin or respiratory tract to maintain homeothermy.

Although sheep have been shown to have a high heat tolerance[1,2], no direct measurements of loss of water from the skin and from the respiratory tract have been reported for this animal. Such measurements have now been made in young lambs subjected to a wide range of environmental temperatures, and the relative importance of the two routes of evaporative heat loss has been assessed.

Four Merino lambs were examined during the first fifteen days of life; they were fasted for 4–12 hr. before each experiment, which lasted 1–1½ hr. The experiments were made in a closed-circuit respiration apparatus (indirect calorimeter) previously outlined[3], but here modified so that respiratory and cutaneous water losses could be determined separately. The lamb, in the chamber of the apparatus, respired into an air-tight polythene mask which covered the head and through which air was circulated. Oxygen was metered into the mask from a spirometer, and expired water vapour and carbon dioxide were determined by weighing. Cutaneous water-loss from the body and limbs was measured by weighing the water absorbed from the air circulating within the chamber. Water vapour pressures at the mask outlet and in the chamber were usually maintained within 3 mm. mercury of each other; they ranged from about 3 mm. mercury at 8° C. to 20 mm. mercury at 45° C. Turbulent air flow over the lamb was at the rate of about 30 cm./sec. Measurement of loss of water, etc., was usually not begun until ¾–1 hr. after the lamb had first been subjected to the experimental conditions.

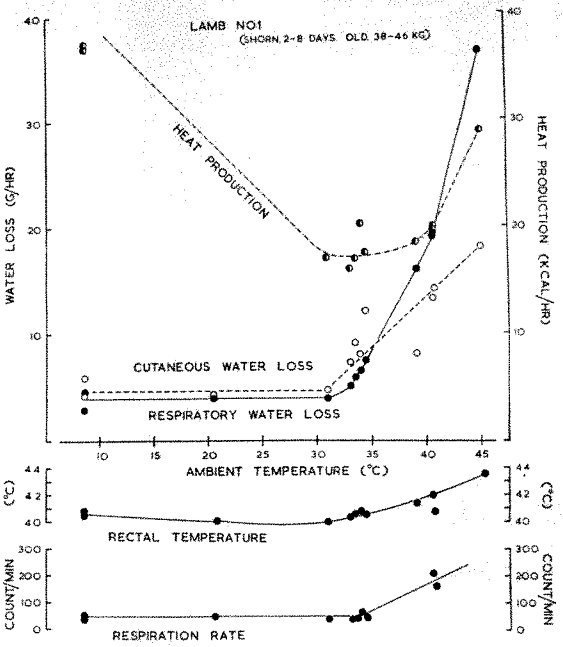

Fig. 1. Heat production, loss of water by evaporation, rectal temperature and respiration-rates in a shorn lamb at various ambient temperatures

The results from lamb No. 1 on which most observations were made are shown in Fig. 1. This lamb had been shorn closely to eliminate errors due to release or absorption of water by the fleece during the experiments. At ambient temperatures below 30° C., losses of water from the skin and from the respiratory tract were relatively constant, the former slightly exceeding the latter. Above 30° C., loss of water from both sources increased with increasing temperature, the rate of increase of the respiratory loss being the greater. Heat production and rectal temperature were minimal at 30° C.; respiration-rate was constant below 30° C. and increased above 30° C. Similar qualitative results were obtained in three other lambs which were not shorn (Fig. 2).

The break in the curve relating ambient temperature to cutaneous water loss suggests that an active

Table 1. Evaporative Water Loss from Lambs with Cutaneous or Respiratory Channel Blocked. Ambient Temperature, 40–41° C.

Lamb	Channel blocked	Water loss (gm./hr.)		Heat production (kcal./hr.)	Rectal temperature (° C.) during experiment	
		Cutaneous	Respiratory		At start	After 1 hr.
No. 1 (shorn)	Neither	14·3	19·5	19·3	40·8	40·8
Age: 8 days	Cutaneous—'completely'	*	32·4	21·9	41·8	41·8
Weight: 4·7 kgm.	Respiratory—partly†	17·2	*	30·7	41·3	43·3
No. 5 (unshorn)	Neither	11·7	36·7	34·3	42·3	42·9
Age: 15 days	Cutaneous—'completely'	*	54·6	42·0	43·7	44·8
Weight: 7·3 kgm.	Respiratory—partly†	11·2	30·6	42·5	42·1	44·9

* Not determined.
† Water vapour pressure was gradually increased from 15 to 55 mm. mercury during the experiment.

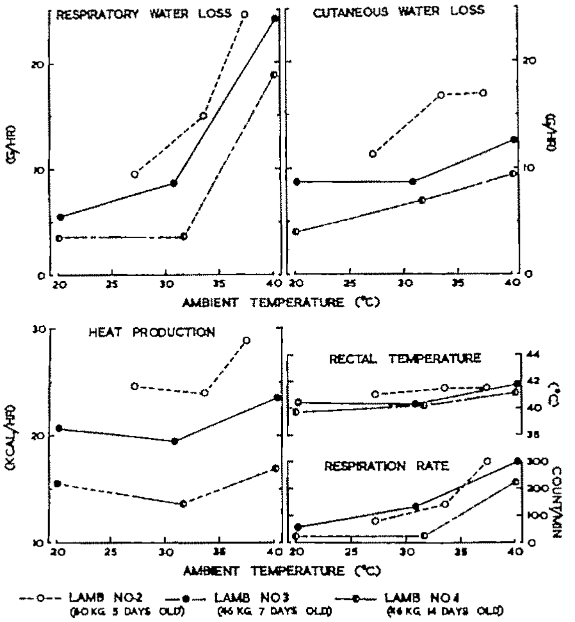

Fig. 2. Heat production, loss of water by evaporation, rectal temperature and respiration-rates in three unshorn lambs at various ambient temperatures

process had come into play above 30° C.; that is, sweating occurred.

The functional reserve of each channel of evaporative cooling was examined in two lambs at 40–41° C. by raising the humidity close to saturation in one channel, to reduce or virtually prevent evaporation, and measuring water loss from the other (Table 1).

In lamb No. 1 when loss of heat by evaporation from the skin was blocked, the loss by respiration was increased to compensate and it was able to prevent a severe rise in rectal temperature. On the other hand, when loss by respiration was much reduced, cutaneous loss was not capable of any marked response and rectal temperature rose rapidly. Although lamb No. 5 was unable to keep its rectal temperature down even when neither channel was blocked, its responses were similar to those of lamb No. 1.

These findings show that, in a hot environment, lambs lose heat by both panting and sweating, and that in the region of minimal heat production the contribution of each is about equal. However, at higher ambient temperatures, the contribution of panting becomes by far the greater, and there is a great functional reserve of evaporative cooling by this route. Sweating appears to approach its limit at that ambient temperature where heat losses by radiation, convection and conduction cease (Table 1).

Published data on adult sheep suggest that they, too, sweat[1,4,5]; but there is insufficient evidence to indicate whether the adult shows the same quantitative relation between sweating and panting as the young. However, results with lambs suggest that in the sheep the relative importance of sweating and panting differs from that in the few other species for which data are available. In the dog[6], sweating is of no consequence, while in the camel[7] and man it is virtually the sole means of evaporative cooling. Cattle, too, appear to rely mainly on sweating[8], but loss of water by respiration shows some increase in heat stress. In sheep, however, panting is apparently the more important mechanism of heat elimination, but sweating can contribute substantially to evaporative cooling.

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[1] Lee, D. H. K., and Robinson, K. W., *Proc. Roy. Soc. Qd.*, **53**, 189 (1941).

[2] Macfarlane, W. V., Morris, R. J. H., and Howard, B., *Aust. J. Agric. Res.*, 9, 217 (1958).

[3] Alexander, G., *Proc. Aust. Soc. Anim. Prod.*, **2**, 10 (1958).

[4] Riek, R. F., Hardy, M. H., Lee, D. H. K., and Carter, H. B., *Aust. J. Agric. Res.*, 1, 217 (1950).

[5] Knapp, B. J., and Robinson, K. W., *Aust. J. Agric. Res.*, **5**, 568 (1954).

[6] Hammel, H. T., Wyndham, C. H., and Hardy, J. D., *Amer. J. Physiol.*, **194**, 99 (1958).

[7] Schmidt-Nielsen, K., Schmidt-Nielsen, B., Jarnum, S. A., and Houpt, T. R., *Amer. J. Physiol.*, **188**, 103 (1957).

[8] Taneja, G. C., *J. Agric. Sci.*, **50**, 73 (1958).

Effect of β-Hydroxybutyrate Injections on the Pancreatic Activity of Guinea Pigs and Rats

A PRODUCT of the intermediary metabolism of fats, namely, β-hydroxybutyric acid, appears in excess during disturbed metabolic processes, especially in diabetic organisms. The accumulation of this intermediary product causes different pathological conditions (acidosis, etc.) in the animal. M. C. Nath and H. D. Brahmachari[1] and H. D. Brahmachari and Mahendra Kumar[2] conclude that chronic administration of β-hydroxybutyrate to guinea pigs and rabbits causes, after a transitory hypoglycæmia, a permanent hyperglycæmia and a lowered glucose tolerance, hypo-insulinism due to atrophy through over-stimulation of the *B*-cells of the pancreas.

Similar observations have been made in our laboratory[3], but after chronic administration of adrenocorticotrophic hormone. We concluded that the parenteral administration of β-hydroxybutyrate produces a mobilization of the adrenocorticotrophin from the hypophysis and, as a consequence, the symptoms observed by Brahmachari *et al.*[2] may be secondary processes due to excess production of corticosteroids as a consequence of the mobilization of adrenocorticotrophic hormone.

We have therefore extended the investigation. Experiments have been carried out on guinea pigs and on white rats of both sexes, divided into four experimental lots, each composed of 5 animals: (1) Normal rats and guinea pigs; (2) hypophysectomized rats; (3) normal rats and guinea pigs treated with β-hydroxybutyrate, 50–100 mgm. per animal; (4) hypophysectomized rats treated with the same dose of β-hydroxybutyrate.

The following determinations were made on each animal: (*a*) extent of glycæmia (photocolorimetrical method, Somogyi–Nelson[4,5]); (*b*) quantitative estimation of plasma-insulin (Vallance-Owen *et al.*[6]); (*c*) estimation of adrenocorticotrophic hormone in blood plasma (Sayers *et al.*[7], modified by us[8]).

The results given in Table 1 demonstrate clearly that during the administration of β-hydroxybutyrate (sodium salt) the glycæmia, after a temporary lowering, rises to 210 mgm. per cent. At the same time the quantity of plasma insulin—after a considerable

Table 1. EFFECT OF THE CHRONIC TREATMENT WITH SODIUM β-HYDROXYBUTYRATE ON NORMAL ANIMALS

No.	Days of injections	Glycæmia (mgm. per cent)	Insulin in plasma (microunits/ml.)	Adrenocorticotrophic hormone in plasma (milliunits/100 ml.)
1	0 (control)	98	92 ± 6	0·5 ± 0·1
2	7	69	310 ± 18	8·5 ± 3·1
3	14	71	360 ± 11	2·1 ± 2 3
4	28	110	135 ± 8	3·3 ± 1·1
5	35	180	60 ± 18	9·3 ± 3 1
6	42	210	80 ± 11	5·8 ± 5·1

increase—decreases to less than the normal values, whereas the level of adrenocorticotrophic hormone from the blood rises to about 10 times that of the initial values, that is to say, the normal values.

We observed in rats treated for 14, 28 and 42 days with this product a destruction of pancreatic *B*-cells, a destruction similar to that observed in rats treated chronically with adrenocorticotrophic hormone, as described by us[8].

These results confirm Brahmachari's observations concerning the diabetogenic effect of the β-hydroxybutyrate.

In order to analyse the mechanism of the action of this substance, we subjected some rats to hypophysectomy, afterwards treating them chronically with this intermediary product. We supposed that if the β-hydroxybutyrate would attack the *B*-cells directly, then even in the absence of the pituitary gland (absence of adrenocorticotrophic hormone) the diabetogenic effect would still take place. But if the pancreas remains intact, that is to say, if the symptoms described above do not appear, then we can assert that this diabetogenic effect of β-hydroxybutyrate is not a primary one, but is mediated by the pituitary gland.

Our results (Table 2) with the hypophysectomized rats after a chronic treatment with sodium β-hydroxybutyrate show an oscillation of the plasma insulin values, but within normal limits. Glycæmia also shows an increase but of little importance. Histologically no destruction of the *B*-cells was observed even after a longer treatment.

We then treated a series of hypophysectomized rats with sodium β-hydroxybutyrate + adrenocorticotrophic hormone (0·5 μgm./100 gm.). In these animals the plasma insulin value decreased, some hyperglycæmia occurred, and the histological aspect of the *B*-cells showed signs of partial atrophy in general. It must be emphasized that though the β-hydroxybutyrate + adrenocorticotrophic hormone, in the absence of hypophysis, provoke diabetic signs, they are less important than those observed in normal animals.

Table 2. EFFECT OF THE CHRONIC TREATMENT WITH SODIUM β-HYDROXYBUTYRATE UPON HYPOPHYSECTOMIZED ANIMALS

No.	Days of injections	Glycæmia (mgm. per cent)	Insulin in plasma (microunits/ml.)	Adrenocorticotrophic hormone in plasma (milliunits/100 ml.)
1	0 (control)	88	44 ± 4	0 0 ± 0 0
2	7	91	66 ± 1 3	—
3	14	90	49 ± 7	—
4	28	118	71 ± 11	—
5	35	101	31 ± 6	—
6	42	87	91 ± 8	—

Table 3. EFFECT OF THE SODIUM β-HYDROXYBUTYRATE + ADRENOCORTICOTROPHIC HORMONE TREATMENT ON HYPOPHYSECTOMIZED ANIMALS

No.	Days of injections	Glycæmia (mgm. per cent)	Insulin in plasma (microunits/ml.)	Adrenocorticotrophic hormone in plasma (milliunits/100 ml.)
1	0 (control)	78	60 ± 10	0 0 ± 0 0
2	7	130	120 ± 21	—
3	14	127	150 ± 11	—
4	28	140	99 ± 0	—
5	35	121	60 ± 8	—
6	42	154	40 ± 1	—

We therefore conclude that: (1) Chronic treatment with sodium β-hydroxybutyrate produces a diabetic state in normal animals, but the effect is not a direct one; it takes place via the hypophysis. (2) The consequences of the chronic treatment with this product are very similar to those obtained by us with a longer treatment with adrenocorticotrophic hormone. (3) The diabetic effect upon the hypophysectomized animals treated with sodium β-hydroxybutyrate has not the importance initially supposed. The sodium β-hydroxybutyrate leads to mobilization of adrenocorticotrophic hormone and there may be hypersecretion of other diabetogenic factors (somatotrophic-hormone); and thus in the absence of the pituitary gland, even in the presence of exogenous adrenocorticotrophic hormone, the diabetic signs are less significant. (4) The diabetogenic effect of the sodium β-hydroxybutyrate as well as of the adrenocorticotrophic hormone are diminished in the absence of the pituitary gland.

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[1] Nath, M. C., and Brahmachari, H. D., *Nature*, 161, 18 (1948).

[2] Brahmachari, H. D., and Mahendra Kumar, *Nature*, 183, 51 (1959).

[3] Schwartz, A., Madar, J., and Kis, Z., *Studii si Cercet. de Medicina Academia RPR. Cluj.*, No. 3–4, 291 (1957).

[4] Somogyi, M., *J. Biol. Chem.*, 160, 61 (1945).

[5] Nelson, N., *J. Biol. Chem.*, 153, 375 (1944).

[6] Vallance-Owen, J., and Hurlock, B., *Lancet*, i, 68 (1954).

[7] Sayers, M. A., and Sayers, G., *Endocrinol.*, 42, 379 (1948).

[8] Schwartz, A., Madar, J., and Kis, Z., *Studii si Cercet. de Biologie Academia RPR. Cluj.*, No. 1–2, 46 (1958).

Vasodepressor Effect of Adenosine Phosphates and 5′-Adenylmethylenediphosphonic Acid, an Analogue of Adenosine Triphosphate

MEMBERS of the adenosine, adenosine monophosphate, adenosine diphosphate and adenosine triphosphate series produce a marked vasodepressor effect in the cat anæsthetized with nembutal. In this series, as the number of phosphate groups in the 5′ position is increased, the vasodepressor effect is also increased[1].

Two general hypotheses as to the order of potency in the members of this series may be considered. In the first, cleavage of the polyphosphate side-chain is postulated with or without transfer of phosphate to a substrate or active site. The high activity of adenosine triphosphate in dilating the coronary arteries in the dog led Winbury *et al.*[2] to suggest that "bond energy may serve to trigger some biochemical process leading to greater dilatation".

Alternative explanations for the mechanism of the action of members of this series, which would not involve 'high-energy' bond cleavage, might be found in the complexing or binding capacity of these

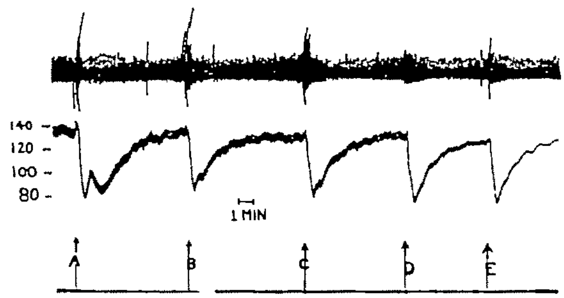
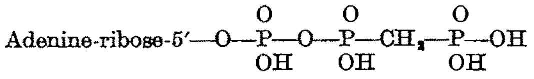


Fig. 1. Respiration (upper) and arterial blood pressure in mm. mercury (lower) tracings from a 2 kgm. cat (male) anæsthetized with nembutal. The letters indicate intravenous injection of the following: (A) 1 µmole analogue; (B) 4 µmoles adenosine monophosphate; (C) 4 µmoles adenosine monophosphate; (D) 2 µmoles adenosine triphosphate; (E) 2 µmoles adenosine triphosphate

compounds. Martell and Schwarzenbach[3] have shown that the association constants of the members of this series, with magnesium and calcium complexes, increase with the number of phosphate groups. Thus the vasodepressor activity appears to parallel the change in association constants of these compounds.

The present preliminary report describes the results obtained in comparing the vasodepressor effect of adenosine monophosphate and adenosine triphosphate with 5′-adenylmethylenediphosphonic acid, a phosphonic acid analogue of adenosine triphosphate (this compound has been prepared and characterized by us; details will be given elsewhere).



$$\text{Adenine-ribose-5}'\text{—O—}\overset{\text{O}}{\underset{\text{OH}}{\text{P}}}\text{—O—}\overset{\text{O}}{\underset{\text{OH}}{\text{P}}}\text{—CH}_2\text{—}\overset{\text{O}}{\underset{\text{OH}}{\text{P}}}\text{—OH}$$

5′-adenylmethylenediphosphonic acid

Experiments were performed on thirty-one cats anæsthetized with nembutal. The carotid blood pressure was recorded with a mercury manometer on a kymograph. Adenosine mono- and tri-phosphate and the analogue were administered intravenously in 0·9 per cent sodium chloride.

Fig. 1 illustrates the results obtained in a cat anæsthetized with nembutal, in which the magnitude of the vasodepressor response is about the same for each of the doses used. The relative potency of the analogue varied from 1·2 to 4·2 times that of adenosine triphosphate in these experiments, with a mean value of 2·3 for 31 such comparisons.

Adenosine monophosphate was found to have a relative potency of 0·22–0·50 that of the triphosphate, with a mean value of 0·33.

The methylene group in place of the pyrophosphate oxygen of adenosine triphosphate results in an increase in vasodepressor activity. This indicates that the terminal pyrophosphate group of the triphosphate is neither necessary for activity nor for enhancement of activity. It is well established that the phosphonic acid (C–P bond) is markedly resistant to chemical cleavage[2,4]. Additional evidence of this resistance to cleavage was seen when the analogue was substituted for adenosine triphosphate in the yeast hexokinase reaction, which reaction has been proved to take place with the cleavage of the adenosine triphosphate terminal pyrophosphate bond[5]. The analogue failed to serve as a substitute for adenosine triphosphate in this reaction. Since the similarity of the pharmacological effects of adenosine triphosphate and its phosphonic acid analogue are striking, this also suggests that cleavage of the terminal pyrophosphate bond is not essential for the production of the vasodepressor effect by adenosine triphosphate. The possibility that the analogue may owe its activity to cleavage of the second pyrophosphate bond, giving rise to adenosine monophosphate and methylenediphosphonic acid, which in turn produces the observed response, seems unlikely since these compounds given together show much less activity than the analogue.

Considering the results reported here, the mechanism of action of the series of compounds may be due to binding to the receptor site without a cleavage of the chain. The activity of the analogue could be accounted for in this manner; that is, binding to the receptor site without cleavage of the chain. The increase in activity of the analogue in comparison with the other compounds may result from a greater affinity for this receptor site.

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[1] Green, H. M., and Stoner, H. B., "Biological Actions of the Adenine Nucleotides" (H. K. Lewis and Co., London, 1950).

[2] Winbury, M. M., Paplerski, D. H., Henmer, M. L., and Hambourger, W. E., *J. Pharm. Exp. Therap.*, 109, 225 (1953).

[3] Martell, E. E., and Schwarzenbach, G., *Helv. Chim. Acta*, 39, 653 (1956).

[4] Kosalopoff, G. M., "Organophosphorus Compounds" (Wiley and Co., New York, 1950).

[5] Colowick, S. P., and Kalckar, H. M., *J. Biol. Chem.*, 148, 117 (1943).

Effect of Different Types of Milk on Intestinal Motility

IN conditions of infantile diarrhœa restriction of milk from the diet causes rapid improvement of the condition: if the milk diet is resumed too early, the symptoms rapidly appear again. This shows that, most probably, there is a relation between the milk and the symptoms of infantile diarrhœa. It may be that in the milk there are factors which stimulate the motility of the small intestine and thus cause its rapid evacuation.

It was found by Becovic and Moglajile[1] that mare's milk contains acetylcholine. This was shown by studying the effect of milk on the isolated frog's heart and the small intestine of the dog prepared *in situ* according to the method of Straub and Triendler. The effects of human milk and the different types of milk obtained from Egyptian dairy animals on the intestinal motility *in vitro* were therefore examined. Fresh milk, stored milk, skimmed milk and pasteurized milk were used.

The jejunum of freshly killed rabbit of about 1 kgm. body-weight was excised. Isolated pieces of jejunum of about 3 cm. in length were suspended in an organ bath of about 10 ml. volume containing oxygenated Tyrode's solution maintained at 37° C. The movements of the longitudinal muscle were recorded by a lever. The normal action and the effect of adding a few drops and 2 c.c. of different types of milk were compared. When the milk showed an excitatory response, its effect was studied on an atropinized loop of intestine.

Fresh milk was collected from the animal by hygienic methods and was immediately examined.

Fresh cow's milk. A few drops had no effect on the intestinal motility. When 2 c.c. of the milk (at 37° C.) were added to the intestinal bath, it caused temporarily slight inhibitory effect after which the intestinal motility gradually increased. Addition of one drop of bellafolin immediately stopped the excitatory effect.

Fresh buffalo's milk. A few drops of the milk added to the intestinal loop caused immediate inhibition of the intestinal motility; this inhibitory effect disappeared very rapidly and the intestine recovered. Addition of 2 c.c. of fresh milk to the intestinal bath caused a marked inhibitory effect, and then the intestine gradually recovered.

Fresh goat's milk. Complete immediate relaxation of the intestinal loop was obtained by the addition of a few drops. A high dose of 2 c.c. produced a similar effect. Recovery gradually occurred.

Fresh human milk. Whether fresh or stored in the refrigerator, human milk had no effect on the motility of the intestinal loop.

The effect of stored milk at room temperature up to 12 hr. or at very low temperature (0° C.) up to 7 days did not cause any change in the behaviour of the intestinal loop.

Pasteurization of milk did not change its effect on the intestinal motility.

The effects of different types of whey were examined.

Cow's whey. Whether in small dose of 5 drops or a big dose of 2 c.c., cow's whey caused a preliminary inhibition followed by strong excitatory effect. One drop of bellafolin immediately stopped the excitatory effect. Cow's whey in high doses did not show the excitatory effect on the atropinized loop.

Buffalo's whey. This produced complete relaxation of the intestinal loop when added even in very small doses.

Goat's whey. The inhibitory effect which was recorded in the case of fresh, stored, and pasteurized goat's milk cannot be shown in case of the whey, but it caused a very strong excitatory effect which is not produced in the case of the atropinized loop.

The preliminary inhibitory effect observed in the case of fresh cow's milk and the maintained inhibitory effect of buffalo's milk were not shown when the milk was previously boiled at atmospheric pressure and then added to the intestinal bath after cooling to 37° C. In the case of goat's milk, boiling did not abolish the inhibition produced.

When the anti-histaminic 'Sandosten' was added to the Tyrode solution in the intestinal bath, the normal effect produced by the different milks were not changed.

The experiments show that different milks have different effects as regards the motility of the intestinal loop *in vitro*. Cow's milk, whether fresh, stored, pasteurized or boiled, had an excitatory effect. The parasympatholytic drug bellafolin abolished this excitatory response. Buffalo's and goat's milk showed a maintained inhibitory effect. As these effects could be observed when the whey was used, the factor concerned with excitation or inhibition of the intestinal loop most probably lies in the whey. Becovic and Moglajile[1] stated that mare's and cow's milk contain acetylcholine, and as the Egyptian cow's milk showed an excitatory effect abolished by bellafolin, so this finding by Becovic and Moglajile can be applied to this milk.

The inhibitory effect produced by goat's milk and buffalo's milk may be due to an inhibitory factor, the nature of which is not yet settled. As the inhibitory effect was abolished by boiling in the case of buffalo's milk, but was not affected by boiling in the case of goat's milk, this indicates that the nature of the inhibitory factor differs in the different milks.

Human milk had no effect at all on the intestinal motility. This may be due either to the absence of such excitatory or inhibitory factors, or they may be present in concentrations antagonizing each other.

The study of the effect of the different amino-acids present in milks and the nature of the excitatory and inhibitory factors is under investigation in our laboratories.

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[1] Becovic, S., and Moglajile, B., *Wiener Tierarztl. Monatsschr.*, 42 (2), 68 (1955).

Hormonal Control of Secretion in the Oviduct of the Amphibia

Galli-Mainini[1], Houssay[2] and Allende[3] have shown that secretion by the glands of the oviduct in *Bufo arenarum* is controlled by a dual mechanism: a prolactin-like hormone from the anterior pituitary acts directly on the oviduct, and a progesterone-like hormone produced in the ovary during ovulation causes secretion independently of the direct pituitary action. In view of the importance from the evolutionary point of view of this demonstration of the action of a progestational hormone at the level of the Amphibia, it was thought desirable to extend the work to include the two British species, *Bufo bufo* and *Rana temporaria*.

In the first series of experiments various concentrations of progesterone ('Progestin' B.D.H.) in 0·2 ml. arachis oil were injected into the lumina of the doubly ligatured left oviducts of ovariectomized frogs and toads[3]. In all experiments using toads the anterior lobe of the pituitary had been removed at least two days previously. 48 hr. after injection the animals were killed, and, after weighing the oviduct and measuring the length enclosed by the two ligatures, the gelatinous secretion within the oviduct was removed and weighed. The results were expressed as the ratio of the weight of secretion in mgm. (after correcting for the weight of oil injected) to the length of oviduct between the ligatures in cm. This expression is termed the quotient of secretion[1,3]. The quotient is numerically smaller in the frog, because, unlike the condition found in the toad, there is no clear differentiation of the oviduct into two parts and it was necessary to use the whole length of the oviduct if the quotients from different animals were to be comparable.

The response to progesterone is shown in Table 1, where a highly significant increase in the quotient of secretion was given by 0·1 mgm. of progesterone.

Table 1. SECRETION BY THE GLANDS OF THE OVIDUCTS OF FROGS AND TOADS IN RESPONSE TO PROGESTERONE OR AN EXTRACT OF OVULATING OVARIES. FIGURES IN BRACKETS DENOTE THE NUMBER OF ANIMALS IN EACH GROUP

Treatment	*Rana temporaria*		*Bufo bufo*	
	Quotient of secretion ± *S.E.*	*P**	Quotient of secretion ± *S.E.*	*P**
0·2 ml. arachis oil in lumen of oviduct	6·07 ± 0·79 (12)	—	12·8 ± 0·996	—
0·5 mgm. progesterone in lumen of oviduct	13·1 ± 3·37 (10)	< 0·03	—	
0·1 mgm. progesterone in lumen of oviduct	12·9 ± 2·36 (7)	< 0·01	40·1 ± 5·32	< 0·001
0·1 mgm. progesterone in dorsal lymph sac	—		41·02 ± 5·18 (9)	< 0·001
0·2 ml. ovary extract in lumen of oviduct	—		29·4 ± 8·11 (8)	< 0·05

* Values of *P* for comparison of experimental groups with arachis oil controls (using right-hand tail '*t*' test).

In the frog increasing the dose to 0·5 mgm. did not appear to promote further secretion. In the toad a very similar response was obtained whether the progesterone was injected directly into the oviduct or into the dorsal lymph sac.

In another series of experiments twelve toads were each injected with 1,000 I.U. of chorionic gonadotrophin ('Pregnyl') and killed 48 hr. later when the animals were in various stages of ovulation. The ovaries were removed, dried at 37° C. for 24 hr., then ground up in a mortar with sand and extracted with ether in a Soxhlet apparatus. 0·2 ml. portions of the clear oil obtained after evaporating off the ether were injected into the ligatured oviducts of ovariectomized and hypophysectomized toads as in the progesterone series. Table 1 shows that this extract gave a higher mean quotient of secretion compared with arachis oil controls, though there was considerable variation in the responses of the assay animals.

These experimental results agree closely with the more detailed work by Houssay and his co-workers on *B. arenarum*, and suggest that the production by the ovary during ovulation of a hormone having a stimulating action on the glands of the oviduct may be a general characteristic of the Amphibia.

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[1] Galli-Mainini, C., *Rev. Soc. Argent. Biol.*, **26**, 166 (1950).

[2] Houssay, B. A., *Rev. Soc. Argent. Biol.*, **26**, 185 (1950).

[3] Allende, I. L. C. de, *C.R. Soc. Biol., Paris*, **130**, 163 (1939).

PLANT PHYSIOLOGY

Response of Growing Plants to a Uniform Daily Rotation

PLANTS under various fixed conditions of light, temperature, and humidity have been turned continuously about a vertical axis at the rate of one revolution a day. The results suggest that the plants used are sensitive to the direction of rotation; a rotation which is clockwise when viewed from above inhibits growth, anti-clockwise direction stimulates growth. As the finding may be useful background to a study of the twining of plants, the initial results are being reported.

Cyclamen. In subdued daylight, or in artificial top lighting, plants which were made to rotate clockwise showed, after two or three days, a loss of turgidity in some of the leaves, which yellowed and died as the experiment continued. Plants turned anti-clockwise were as healthy and turgid as stationary ones.

Scarlet Runner Beans. Beans which normally twine anti-clockwise were found not to twine around their supports when growing plants and their supports were rotated anti-clockwise at one revolution a day under fixed side-lighting with tungsten lamps. Plants which were turned clockwise began to twine but lost turgidity, and some leaves yellowed and died. When these plants were transferred at an early stage of reaction to anti-clockwise movement they showed partial or full recovery.

Oats. Husked oats (variety Milford), were pre-chilled in tap-water at 5° C. for 48 hr. The seed was then sown, groove side down, on moist filter paper in a covered dish, and held at 20° C. in darkness. After 24 hr., germinated seedlings were transplanted on to 4-in. diameter perforated plastic holders over tap-water. Each holder and dish (48 seedlings per holder) was transferred to a completely dark, controlled-climate cabinet provided with a suitable set of turntables to give clockwise and anti-clockwise rotations. Cabinet temperature was controlled to 1° C. and relative humidity to 1 per cent. In successive experiments the controlled temperatures ranged from 25° C. to 28° C. and the relative humidity 85–95 per cent. After 5 days, the lengths of coleoptiles and roots were measured. In every experiment the mean lengths under each treatment were expressed as a percentage deviation from the mean of the stationary plants. The average deviations throughout nine experiments were as follows:

Clockwise	Coleoptiles	− 6·1	per cent
	Roots	− 8·6	,,
Anti-clockwise	Coleoptiles	+ 5·6	,,
	Roots	+ 10·2	,,

In individual experiments the means of the clockwise-rotated sets of plants were always smaller than those of the stationary, and the anti-clockwise in all cases larger (for example, for coleoptiles—clockwise 42·7 ± 0·8 mm., stationary 46·3 ± 0·7 mm., anti-clockwise 48·2 ± 0·9 mm.).

The subject of diurnal rhythms as affecting the above is being considered.

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Energy-trapping Centres in Photosynthesis

IN opaque specimens of packed *Chlorella* cells, the quantum yield of red fluorescence varies with the wave-length of the exciting light[1]. From a maximum value measured at the absorption peak at wave-length 675 mμ, decreases to about 70 per cent of this value occur at shorter wave-lengths, as shown in Fig. 1. In contrast, dilute suspensions with measured true absorption give a relatively constant yield, suggesting that competitive selective scattering and re-absorption losses are largely responsible for the yield decreases observed in opaque specimens.

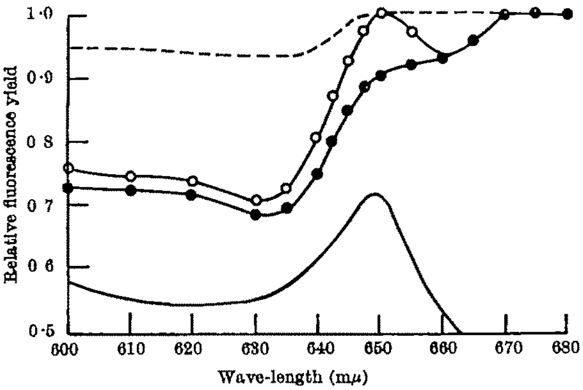

Fig. 1. Dependence of the relative fluorescence on the excitation wave-length and intensity in *Chlorella*. – – – –, Thin suspension; ○——○, opaque specimen (high intensity); ●——●, thick specimen (low intensity), ———, difference spectrum produced by intensity (ordinate × 2)

It has now been found that a part of the effect depends on the light intensity employed in yield determinations. High-intensity light produces a relatively more constant yield compared with low intensities, and the spectral dependence of the relative yields of *Chlorella* at different levels of illumination are included in Fig. 1. The absolute fluorescence yield at high-intensity excitation was 3–4 times greater than at low light levels. Since no changes in absorption or fluorescence spectra larger than 1 per cent occurred during these measurements, the difference spectrum corresponding to the selective increase in yield must be produced by a change in the effective fluorescence yield of a component pigment. The similarity in position of the difference spectrum maximum, shown in Fig. 1, to that of chlorophyll *b* *in vivo* suggests the involvement of this pigment. Before discussing the significance of this observation, it is necessary to consider the different situations of chlorophylls *a* and *b* in relation to energy transfer. Assuming that this process is responsible for energy migration to photochemically active trapping centres present in relatively small concentration, then these centres must contain a non-fluorescent pigment with an absorption band overlapping the fluorescence band of chlorophyll *a*. Changes in the trapping efficiency, accompanied by inverse changes in fluorescence yield, can occur in two ways if the geometry of the system remains constant. A change in absorption in the trapping pigment could reduce energy acceptance, or alternatively, the trapping pigment can become fluorescent. The absence of considerable absorption and fluorescence spectral changes means either that the trapping pigment is present in such small concentration that its absorption changes are undetected or that it is chlorophyll *a* itself or a spectroscopically similar molecule. The reported similarity of the fluorescence and delayed-light spectra[2] observed in *Chlorella* supports the involvement of chlorophyll *a*. On this latter basis, the simplest model for a trapping centre would be a complex between chlorophyll *a* and a colourless acceptor molecule, capable in its ground-state of quenching the chlorophyll singlet excited state, being itself converted to a metastable, chemically active form. Providing the ground-state interaction of the components is small, the associated spectral changes will be also small, as they are in chlorophyll *a* quenching by complex formation with aromatic nitrocompounds and quinones[3].

Detectable spectral shifts occur with chlorophyll *b* however, and some *in vivo* measurements of absorption changes in illuminated *Chlorella*[4] may be relevant in this connexion. During the life-time of the metastable form, the associated chlorophyll molecule could fluoresce normally. Calculation indicates that about 3 per cent of the total chlorophyll, complexed in trapping centres, would receive by resonance transfer at least 75 per cent of all absorbed quanta, the balance appearing as heat or fluorescence. In the trapping centres described, chlorophyll *b* might replace chlorophyll *a* in some instances. Few transfers among chlorophyll *b* molecules can occur *in vivo*; because of the relatively lower concentration of this pigment, and the large overlap of its fluorescence band with the absorption peak of chlorophyll *a*, excited chlorophyll *b* always transfers directly to chlorophyll *a*. As a consequence, that fraction of chlorophyll *b* which does not sensitize fluorescence must consist entirely of quenched molecules. This fraction corresponds to about 10 per cent of the whole absorption at 650 mμ, or to about 25 per cent of all chlorophyll *b*, which contributes roughly 40 per cent to the total absorption at this wave-length. The transfer processes occurring between the chlorophyll types can be shown in the following scheme:

$$\begin{tikzcd}
b \arrow[rr] & & {[a]} \arrow[rr] \arrow[dr, "\bar{n}"] & & \text{Fluorescence} \\
& b.T^{*} \arrow[ur] & & a.T \to \text{Photosynthesis} & \\
& & b.T \to \text{Photosynthesis?} & &
\end{tikzcd}$$

where T and T^{*} represent the acceptor molecule in the ground and metastable states, respectively, and $\bar{n}$ is the average number of transfers among chlorophyll *a* molecules before trapping. A feature of this scheme is that at high intensities of excitation both types of chlorophyll *b* molecules sensitize chlorophyll *a* fluorescence and photosynthesis. The negligible overlap of chlorophyll *a* fluorescence with chlorophyll *b* absorption prevents the quenched fraction of this pigment from trapping chlorophyll *a* fluorescence. If this overlap is not negligible, however, a scheme requiring only chlorophyll *b* trapping centres could be proposed. The possibility that the chlorophyll *a* and *b* trapping centres may be shared or photochemically different is suggested by recent reports of the effect of supplementary light on photosynthesis[5]. In addition, the fall in photosynthetic yield in *Chlorella* which is observed at certain wave-lengths[6] and is usually attributed to inactive carotenoid absorption, could equally well be attributed to an inactive fraction of chlorophyll *b*, as was indeed suggested by Emerson.

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[1] Duysens, L. N. M., thesis, University of Utrecht (1952).
[2] Arnold, W., and Davidson, J. B., *J. Gen. Physiol.*, **37**, 677 (1954).
[3] Livingston, R., and Ke, C.-L., *J. Amer. Chem. Soc.*, **72**, 909 (1950).
[4] Strehler, B. L., and Lynch, V., *Arch. Biochem. Biophys.*, **70**, 527. (1957).
[5] Emerson, R., and Chalmers, R. F., *Phycol. Soc. Amer.*, **11**, 51 (1958).
[6] Emerson, R., and Lewis, C. M., *Amer. J. Bot.*, **30**, 165 (1943).

BIOLOGY

An Automatic Fish Egg Counter

The fecundity of fish has hitherto been generally determined by making direct counts of weight or volume sub-samples of the total gonad contents. These counts are then raised by the ratio of the total weight or volume of the gonad contents to the sample weight or volume.

This process is very time-consuming, and, in consequence, routine studies of the fecundity of large numbers of fish have not been practicable. While typical fecundity estimates have been obtained for a large number of fish species, using material classified by size and age, and collected in one or two seasons, no detailed long-term investigations have been conducted. Such investigations could be of considerable value in studies of the biology and population dynamics of fish populations where it is desired to know the changes in fecundity from year to year, the factors determining these changes, and the dependence of recruitment and strength of year-class on these changes.

To meet this need for routine measurement of fecundity (in the first instance of herring) an apparatus has been devised here. This makes rapid counts of all the eggs in a gonad, thus eliminating the tedious work of hand-counting, and also eliminating the errors of sub-sampling.

The apparatus is shown diagrammatically in Fig. 1. This shows that the eggs are drawn in a stream of water through a constricted tube, interrupting a beam of light which is directed through it. The light beam falls on the cathode of a photomultiplier tube, and the interruptions of the beam are recorded on an electronic counter.

The constricted tube, or counting chamber, has a minimum internal diameter of 1·53 mm., and it has been found that this is just large enough to pass the largest herring eggs without causing frequent stoppages.

The photomultiplier tube used is an 'R.C.A. type 931*A*' with nine dynodes, and the light source is a 2·2-V. 0·25-amp. lens-end flash-light bulb. The counter is a 'Panax type *D*554' which conveniently supplies the E.H.T. for the photomultiplier (roughly 200 μamp. at 1,000 volts) and an a.c. supply for the light source.

As will be seen from Fig. 1, a flow of bubble-free water is maintained from a constant-head tank through the counting chamber. Eggs to be counted are first cleared of all connective tissue and placed in water in the large glass funnel, and the stirrer speed is adjusted to bring them into suspension. The tap at the base of the funnel is then opened, and the eggs sink slowly downward and are swept through the counting chamber. The rate of sinking, and so the counting-rate, can be adjusted by varying the speed of the stirrer. In practice, a counting-rate of about 100 per sec. is found to be best. Attempts to increase this rate generally result in blockage of the system.

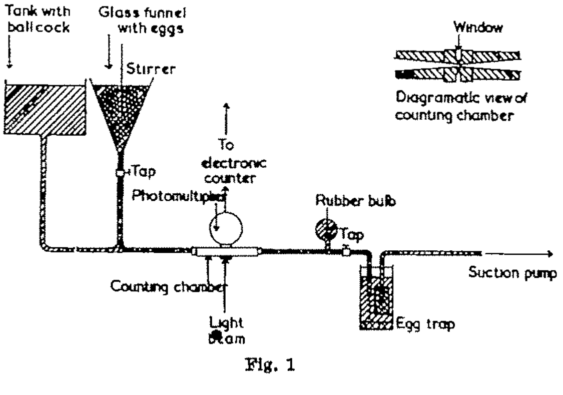

Fig. 1

A considerable number of trials have been made to gauge the accuracy and consistency of the counts. Typical sets of results are shown in Table 1, which gives small successive counts of samples of known size, and in Table 2, which shows successive counts of large samples.

Table 1

Sample	Actual number of eggs	No. recorded by counter in successive trials			
		1	2	3	4
1	521	521	519	525	523
2	778	775	777	771	770
3	507	505	505	505	503
4	808	805	803	801	800

Table 2

Sample	No. of eggs recorded by counter		
	1	2	3
1	19,088	19,128	19,101
2	25,093	25,135	25,274
3	39,494	39,513	39,596
4	43,106	43,150	43,279

These results show that the maximum discrepancy between counts and the maximum error on known samples does not exceed 1 per cent. It is thus felt that reliance can be placed on the method, and a programme of routine investigations of herring fecundity is being developed accordingly.

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A Pale Hag-fish

De Graaf[1] and more recently Ewer[2] have described specimens of *Xenopus laevis* lacking hæmoglobin. In the course of an investigation on the physiology of the heart of the cyclostome *Myxine glutinosa*, I found on August 20 an animal about 25 cm. long which was remarkably pale, except in the caudal and dorsal regions, which were grey. Its blood appeared to have no pigmentation due to hæmoglobin except near the gills, where there were some small localized red areas. All the blood vessels were cream in colour as well as the heart, although the ventricle might have been pink (? myoglobin, cytochromes). The liver was dark green and after fixation in 8 per cent formol the principal dorsal vessels showed a light greenish coloration due perhaps to the destruction of hæmin compounds. As for the gall bladder, it did not appear greener than normal.

When first observed, the animal was in the dark in circulating sea water at 15° C. among a mixture of hag-fish, and showing as much vitality apparently as the other animals; it is particularly interesting to emphasize that this specimen had been caught at the same time as the others, two days before, in the Gullmars Fjord, Sweden, using G. Gustafson's technique, that is, by means of a large bottle containing some pieces of mackerel. Thus this animal, which is very poor in respiratory blood pigments, shows normal habits of feeding, proving that its olfactory organs were in good physiological condition. Hence the pathological state of the blood of this hag-fish leads one to ask what is the limiting role which could be taken by hæmoglobin in the life of *Myxine*.

My thanks are due to Dr. Swedmark, director of the Kristinebergs Zoologiska Station, Fiskebäckskill, Sweden, for providing many living hag-fishes.

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[1] De Graaf, A. R., *J. Exp. Biol.*, 34, 178 (1957).

[2] Ewer, D. W., *Nature*, 183, 271 (1959).

[3] Gustafson, G. J., *Ark. Zool.*, 28, A, No. 2 (1934).

Interlaminate Figures of Sessile Barnacle Shells

Alessandri[1], Gruvel[2] and Davadie-Suaudeau[3], in their works on the cirripedes, have directed attention to figures or patterns in the shell plates of various species of sessile barnacles, and further work may prove these to be an important taxonomic characteristic for species identification; but this seems to have been somewhat neglected.

The usual method of identifying barnacles is often difficult due to the variability of the shell configurations and must frequently be followed by examination of the soft parts—a tedious undertaking. Even then, mistakes in naming have occurred, but work by Cornwall[4-6] on many specimens has shown that the 'interlaminate' figures are specific and constant for recent and fossil barnacles.

The following method of revealing the figures is a modification of the procedure using plaster of Paris as described by Cornwall. Whole specimens or plate fragments are embedded in 'N.H.P.' mounting plastic (obtained from North Hill Plastics, Ltd., London). Rings of tubular rubber, polythene or cardboard are cut to fit the size of the specimen to be mounted. The rings are placed on a glass plate to which is applied a thin coating of 'Vaseline' and the specimen carefully placed, basis downwards, within the ring. A small amount of the plastic powder ('N.H.P. 2031/19') is spread over and around the specimen and thoroughly wetted with the plastic fluid ('N.H.P. 1844'). When the first application of liquid has been absorbed a fresh portion of powder, followed by more liquid, is added. This procedure is repeated until all, or as much as is thought necessary, of the specimen is covered. Leave to dry and harden (30–60 min.). When complete specimens are to be mounted it is advisable to fill the shell cavity with plastic by turning the mounting ring over and repeating the above process. When ready, the embedded specimen is extruded from the ring or, if this proves impossible, it is cut away and is ready for grinding and polishing.

It is necessary, in order to show the figures to their best advantage, to ensure that the surface to be examined is as flat and even as possible. A coarse grinding is done on a glass plate with wet 320 silicon carbide powder followed by a similar grinding on a second plate with 700 powder. Grinding should be done until the parietal canals are visible. This can be controlled by examining the ground surface at intervals with a hand lens. After grinding, the surface is given a rinse with water to remove any adhering carbide particles and then dried by pressing on to a filter paper. Polishing is done with diamond paste on a chamois leather followed by a final buffing on a dry chamois leather.

The figures are best seen at a magnification of between 20 and 50 with the light from the top and slightly to the side of the mounted specimen. A few drops of water or benzyl alcohol[5,6] run on to the prepared surface between a cover-slip will often help reveal figures which tend to 'disappear' when the mount is dry. Orienting the specimen to the light is also a help in showing up details of the figures. One of the great advantages of the method outlined above is that the mounted specimen can be dissolved out of the plastic mount by steeping in chloroform, when it can be retrieved intact except for the small portion which has been ground away.

Certain species such as *Balanus balanoides* and *Elminius modestus* have not shown any figures in preparations from the basis. This may be due to the fact that these species do not have any parietal canals (Crisp, D. J., personal communication). It may be that such species will show figures if preparations are made from the ala of the wall plates[6].

An atlas of the interlaminate figures from identified barnacle specimens would be of great value for the further identification of species and would help in furthering this work.

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Dec. 12.

[1] Alessandri, Guido de, *Boll. Soc. Geol. Italiana*, 13, 3, 234 (1895).

[2] Gruvel, A., "Monographie des Cirrhipedes ou Theocostraces" (1905).

[3] Davadie-Suaudeau, (Madam) Claude, Bull. du Service de la Carte Géologique de l'Algérie, Ser. 1, Paléontologie, No. 14 (1952).

[4] Cornwall, I. E., *J. Paleontol.*, 30, 3 (1956).

[5] Cornwall, I. E., *Canad. J. Zool.*, 36, 79 (1958).

[6] Cornwall, I. E., *Canad. J. Zool.*, 37, 401 (1959).

Seed Incompatibility in *Primula*

When diploid and autotetraploid plants of *Primula veris* are artificially crossed reciprocally, fertilization occurs and seed is set, but the seed is bad and germination is nil; the same is true for *Primula vulgaris* and *P. elatior*. Similar seed incompatibility (as it may be called) between diploids and their colchicine-induced autotetraploids has been reported in several other genera of flowering plants, for example, *Gossypium*[1] and *Galeopsis*[2]; it is associated with post-fertilization breakdown of the seed tissues, in particular the endosperm. On the other hand, in some genera, for example, *Beta*[3], the crosses are easily made and viable triploids can be produced in large numbers. The reasons for these different types of behaviour are not known, but they are probably connected with differences in the nature and mode of functioning of the tissues concerned in the nutrition and protection of the embryo; it is worth noting that these tissues (the endosperm and the maternal nucellus and endothelium) generally differ in genetic constitution both from the embryo and also from one another.

The seeds produced in the reciprocal diploid–tetraploid crosses in *Primula* are of two types, according to the direction of the cross; similar contrasting seed types are found in a number of diploid interspecific crosses in this group[4]. This is shown in Table 1, where the *P. elatior–P. veris* cross is used as an illustration.

In these crosses, type *A* seed is very small, with abnormally thick integuments, a small amount of

Table 1

♀ parent		♂ parent	Seed type
P. veris (2*n*)	×	*P. veris* (4*n*)	*B*
P. elatior (2*n*)	×	*P. veris* (2*n*)	*B*
P. veris (4*n*)	×	*P. veris* (2*n*)	*A*
P. veris (2*n*)	×	*P. elatior* (2*n*)	*A*

endosperm, which usually degenerates, and a small, often persistent embryo. Type *B* seed is of normal size; the integuments are thin, and the endosperm and the minute embryo degenerate soon after fertilization. Both types of seed are usually nearly empty at maturity and often the seed walls collapse; (very rarely a viable diploid hybrid may be produced from the *veris–elatior* (type *A*) cross[5]). The classification of the seeds into types *A* and *B*, originally based on seed dissection, is fully supported by detailed embryological investigation of their development and breakdown; so that the two pairs of crosses described in Table 1 may be regarded as analogous. In one, seed incompatibility is presumably conditioned by the qualitative gene differences between the two diploid species, and in the other by the quantitative gene differences between the diploid and the autotetraploid.

In previous work on the diploid crosses[4], this analogy led us, following the work of Stephens on *Gossypium*[1] and Howard on *Nasturtium*[6], to think of the genomes of the species in terms of 'genetic strength', and to assign to each genome a numerical value, the genetic value. The results of reciprocal interspecific crosses could then be interpreted in terms of the ratio of the genetic values of the various tissues of the seed, these ratios being different in any one pair of reciprocal crosses. The ratio of the genetic value of the endosperm to that of the maternal tissues has in fact been thus used in interpreting segregation, in interspecific hybrids, of the genes controlling seed incompatibility[7]. In this interpretation, the genetic value of *P. elatior* was set at unity and that of *P. veris* at 1·8, to conform with the fact that the degree of incompatibility in the *elatior–veris* crosses was slightly less than that in diploid–autotetraploid crosses; in the latter, it is assumed that the ratio of the genetic values of the parents is always 1 : 2.

The analogy between the two types of cross can be tested by making reciprocal crosses between the autotetraploid *P. elatior* with a genetic value of 2, and diploid *P. veris* with a genetic value of 1·8. The values of the parents are thus approximately equal, so that theoretically the cross should produce good seed and viable triploid hybrids in either direction; the seeds should tend to type *A* when *P. elatior* (4*n*) is the female parent and to type *B* in the reciprocal. In fact the cross *P. veris* (2*n*) ♀ × *P. elatior* (4*n*) ♂ gives a fair yield of good seeds; germination percentages up to 67 per cent have been recorded, and the hybrids are triploid. Embryological study shows that these seeds are, as expected, of a moderate type *B*; there is a fair amount of endosperm, which shows some abnormalities. On the other hand, the reciprocal cross, *P. elatior* (4*n*) ♀ × *P. veris* (2*n*) ♂, produces rather small seeds, practically empty at maturity, which embryological investigation shows to be of an unexpectedly extreme type *A*, with very thick integuments; and no viable hybrids are produced.

It may be concluded that some at least of the genetical differences which control seed incompatibility between *P. elatior* and *P. veris* are analogous to those between diploids and autotetraploids, and are of a quantitative nature; thus they may be duplications, either of genes or of small chromosome segments. Over and above these differences, however, are others of a qualitative kind; the fact that the reciprocal crosses between *P. elatior* (4*n*) and *P. veris* (2*n*) give such strikingly different results, even though their genetic values are approximately equal, suggests that the qualitative genetic differences may have a cytoplasmic as well as a nuclear component.

A full account of this work is in preparation.

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[1] Stephens, S. G., *J. Genet.*, 44, 272 (1942).
[2] Håkansson, A., *Hereditas*, 38, 425 (1952).
[3] Rasmusson, J., *Nord. Jordbrugsforsk.* (1948).
[4] Valentine, D. H., *Symp. Soc. Exp. Biol.*, 7, 146 (1953).
[5] Valentine, D. H., *New Phytol.*, 50, 383 (1952).
[6] Howard, H. W., *J. Genet.*, 48, 111 (1947).
[7] Valentine, D. H., *New Phytol.*, 55, 289 (1956).

Induction of Traps of Nematophagous Fungi using *Panagrellus redivivus*

FOLLOWING the revived interest in biological control of plant-parasitic nematodes through use of nematode-trapping fungi, much attention is being devoted to collection, identification and economic utilization of these ubiquitous microfloræ.

One conventional method of isolating these forms involves addition of suitable mycogenous material to maizemeal agar. Following a suitable period of growth, during which time fruiting structures are produced, nematode-trapping devices such as adhesive mycelial meshes or knobs, and constricting rings may be in evidence. In such instances the fungus either has been able to form such traps exclusive of the presence of nematodes or else a suitable number of free-living nematodes were present in the mycogenous material and thus stimulated trap formation. In certain cases, however, an external source of nematodes must be utilized in order to provide sufficient impetus for these fungi to exhibit their predacious devices.

Drechsler[1] used additional putrescent material such as friable leaf mould for supplying nematodes to cultures. Comandon and de Fonbrune[2] used sterilized free-living soil nematodes, while other workers employed animal parasitic nematodes[3,4]. Plant-parasitic eelworms such as *Heterodera rostochiensis*[5] and *Ditylenchus dipsaci*[6] also have been used successfully. Duddington[7] found that enormous numbers of free-living nematodes could be reared on rabbit dung infusion agar, but admitted that "One of the major difficulties in working with predacious fungi is maintenance of adequate stocks of eelworms for use when they are required"[8].

During the past two years the nematode *Panagrellus redivivus* (L.) Goodey, 1945, has been utilized with great success for producing fungal traps in cultured nematophagous hyphomycetes. Following growth of moulds from suitable mycogenous material implanted on maizemeal agar, either a water suspension or a writhing mass of *Panagrellus* is introduced into agar plates. Within a day or two fungal trapping structures usually are manifested in legion. Since reproduction of this nematode is continual and prolific, worms of many sizes are produced, allowing even the smallest of traps to become functional. An adequate supply of eelworms can be maintained constantly on cellulose sponges drenched with potato broth[9] or by use of the oatmeal culture method of Cheo and Tarjan[10]. The latter method, which I have used successfully for several years, involves culture of the worms in an oatmeal – cation exchange resin medium. Once the animals have multiplied sufficiently they will form thickly populated, culture medium-free, curd-like masses on sides of the culture vessel above the medium, which can be readily scraped off for introduction into agar plates. Oatmeal cultures in which worms are established can be stored at 2–5° C. When needed, cultures are warmed to room temperature, resulting in accumulation of nematodes on sides of the culture vessel as referred to above. *Panagrellus* populations can be maintained for years by refrigeration, after establishment on test-tube agar slants.

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Oct. 30.

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[1] Drechsler, C., *Biol. Rev.*, 16, 269 (1941).

[2] Comandon, J., and de Fonbrune, P., *C.R. Soc. Biol., Paris*, 129, 619 (1938).

[3] Deschiens, R., *Bull. Soc. Path. Exot.*, 32, 394 (1939).

[4] Schmidt-Hoensdorf, F., and Ehrentreich, F., *Ztrlbl. Vet.*, 4, 390 (1957).

[5] Hutchinson, S. A., and Mai, W. F., *U.S. Dept. Agric. Plants Dis. Rpt.*, 38, 185 (1954).

[6] Duddington, C. L., "The Friendly Fungi", 178 (Faber and Faber, London, 1956).

[7] Duddington, C. L., *Trans. Brit. Mycol. Soc.*, 38, 100 (1955).

[8] Duddington, C. L., *Bot. Rev.*, 21, 432 (1955).

[9] Taylor, A. L., Feldmesser, J., and Feder, W. A., *U.S. Dept. Agric. Plant Dis. Rpt.*, 41, 527 (1958).

[10] Cheo, P. C., and Tarjan, A. C., *Phytopath.*, 45, 184 (1955).

A Mating-type System in an Acellular Slime-mould

An investigation of the life-cycle of the acellular slime-mould (myxomycete) *Physarum polycephalum* has revealed that a mating-type system exists in this organism, and analysis of this system has been carried out. The existence of mating-types in myxomycetes has previously been suggested by Skupienski[1] as a result of his work on *Didymium nigripes* and *D. difforme*, but the situation was not fully investigated.

The spores of *Physarum polycephalum* release uninucleate amœbæ which, after a variable number of divisions, fuse in some unknown way to form the acellular, multinucleate plasmodium. The plasmodium increases in size and, under suitable conditions, produces spores.

A plasmodium, kindly supplied by Dr. Rusch of the University of Wisconsin, produced spores from which a series of cultures was started. Ten out of twenty-five such cultures grown from groups of spores produced plasmodia, but none of twenty cultures grown from single spores did so. These results are similar to those of Skupienski with *D. difforme*.

Groups of amœbæ from ten single isolated spores were used to set up 55 different cultures: 10 of these cultures were started with amœbæ of the ten groups taken separately, and forty-five started with amœb from all possible combinations of the ten grou taken two at a time. The incidence of plasmodiu formation among these cultures was plotted on chart, and the results indicated that a simple tw type (+/−) mating system was in operation (Fig. The test was repeated twice, and in the combin results of all three tests, plasmodium formati occurred in twenty-one of the twenty-five cultur in which it was expected on the basis of a +/ system, but not in any of the twenty combined-gro cultures in which it was not expected or in any of t ten single-group cultures. The failure to for plasmodia in four of the cultures in which it w expected could be due to factors other than matin type, since it was noticed in all experiments th plasmodium formation is dependent also upon number of external conditions, particularly humidit

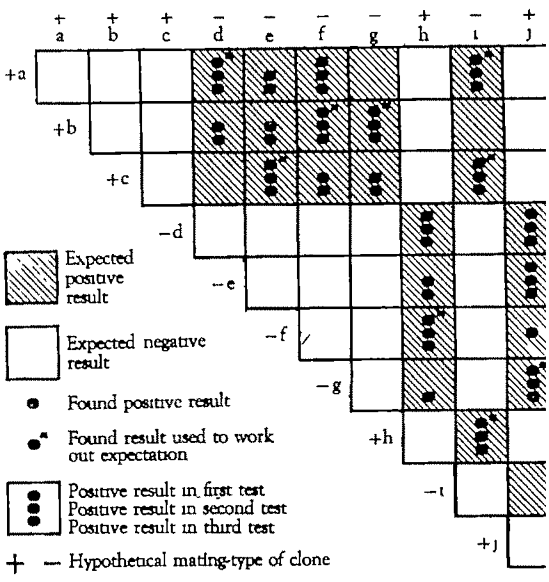

Fig. 1. Tests of the mating-type system. The ten clones were denoted by the letters *a–j*. Each square in the chart represents a culture. Forty-five cultures were started with amœbæ of the ten clones taken in all possible combinations of two at a time, and ten control cultures were started with amœbæ of the ten clones taken separately. The mating-type + or − was assigned to each clone on the basis of nine of the results of the first test. These nine results are marked as shown in the key

It may be concluded from these results that plasm dium formation takes place only when amœbæ different mating-type meet, that only two matin types occur in the cultures investigated, and that spore produces a clone of amœbæ of one mating-ty only.

Investigation of the inheritance of the mating-typ gave the following results. Fifty clones of amœb were established, each derived from a single spore a plasmodium produced by one of the combinatio of two clones described above. Each of these fif first-generation clones was then tested for plasmodiu formation with each of the two parent clones. Fifte of the first-generation clones formed plasmodia wi one parent clone only, ten with the other pare clone only, and twenty-five with neither parei These results are consistent with the hypothesis a simple +/− system. The first-generation clon tested two at a time among themselves gave furth results also consistent with this hypothesis. Howev many of the combinations have failed to produ plasmodia in repeated tests. This suggests, as could be expected, that inherent factors other than mating-type may affect mating and plasmodium formation. While the results are consistent with a two-type mating system, they are not consistent with any of the multifactorial mating-type systems well known in other micro-organisms.

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[1] Skupienski, M. F. X., *C.R. Acad. Sci., Paris*, **167**, 31 (1918); **182**, 150 (1926).

MICROBIOLOGY

Mode of Action of Hair-loosening Organisms

CONSIDERABLE research has been devoted to the development of enzymes for replacing the normal putrefactive techniques for the loosening of wool and hair fibres on animal pelts. Detailed knowledge of the tissues responsible for anchoring the fibre in the skin is therefore essential.

Examination of fibres obtained from skins which have been exposed to bacterial attack shows that the follicle bulb has been completely digested, leaving a tapered root of exposed cortical cells. It is considered that digestion of the root bulb permits the fibre to be readily withdrawn from the skin and that the organisms responsible penetrate down the follicle to attack the soft keratin.

In recent work, however, we have related the appearance of the follicle to the force necessary to withdraw fibres from pelts and have observed that, when the depilation load has fallen to the level at which the fleece is normally pulled from the skin, the whole follicle is removed intact with the fibre. At a later stage of putrefaction, although the depilation load is virtually unchanged, the follicle is rapidly digested to give the typical tapered appearance.

Fig. 1 shows a bundle of intact follicles on fibres pulled as soon as the depilation load had fallen to a level suitable for pulling.

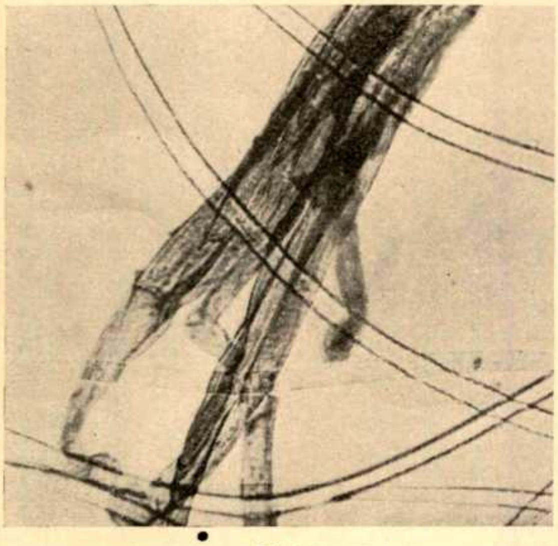

Fig. 1

It seems, therefore, that the bacteria penetrate from the flesh side of the skin to proliferate around the follicles, and that the most significant feature of fibre loosening is the digestion of material in these regions. Whereas efforts have, in the past, been concentrated on the degradation of the soft keratin, the production of an enzyme designed to digest the dermal material immediately adjacent to the follicle would seem likely to result in a greater yield of undamaged fibre.

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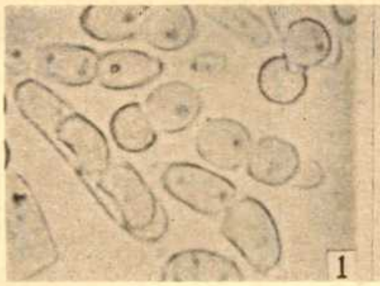
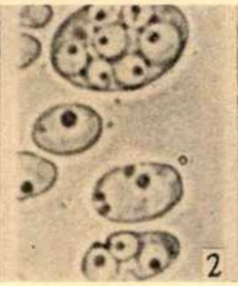
Taxonomy of Yeasts

ON different occasions, when studying the classification of yeasts, I have been able to appreciate how little consistency there is in the taxonomic characters in use. Recently, I have met with fresh anomalies which, in my opinion, make a complete revision of the present system of classification of yeasts unavoidable.

(*a*) From the aqueous solution separated from oil during the manufacture of olive oil, in Spain named 'alpechín', a yeast culture was isolated, *Ac* 11, which is a characteristic *Zygosaccharomyces*, able to produce vigorous alcoholic fermentation and which ferments glucose, galactose, saccharose, melibiose and raffinose completely, and does not ferment or assimilate maltose or lactose. In accordance with the classification of Lodder and Kreger-van Rij[1], this yeast could be considered as a *Sacch. microellipsodes*; however, as to the organism described by Lodder and Kreger-van Rij as a *Sacch. microellipsodes*, is it truly a *Saccharomyces*? The Dutch authors admit that they have not been able to observe spores in the strain under study, and neither could Stelling-Dekker in 1931; moreover, Kudriavtzev[2] categorically denies that the organism is a *Saccharomyces*, not because of its inability to sporulate, but because of its morphology, its weak fermentation of the carbohydrates, etc.; and states that, in his opinion, its place in the classification system is not yet sufficiently clear. Therefore, it is possible that our organism *Ac* 11 corresponds to a *Saccharomyces* species which has not yet been described, for which I propose the name of *Sacch. malacitensis* (from Málaga, the province from which I received the sample of 'alpechín').

Moreover, I had earlier obtained a hybrid[3] crossing *Sacch. oxidans* (ferments glucose, saccharose and raffinose 1/3) and *Sacch. oleaceus* (ferments glucose, galactose, melibiose and raffinose 1/3), to which I gave the symbol *H* 7–4–1; this has a fermentation spectrum identical with that of *Sacch. malacitensis*. Though there are notable differences between the morphology of *H* 7–4–1 and *Sacch. malacitensis*, it would be reasonable to suppose that, were the origin of *H* 7–4–1 unknown, it would be classified as a *Sacch. malacitensis* strain. However, in biology it is inconsistent to consider as a new species an organism obtained by hybridation with other diverse organisms, since a species includes all the potentially interbreeding individuals, the descendants of which are also potentially interbreeding. Therefore the doubt arises whether the present separation of species in the genus *Saccharomyces* is admissible, based as it is fundamentally on the simple difference of fermentation of only one carbohydrate.





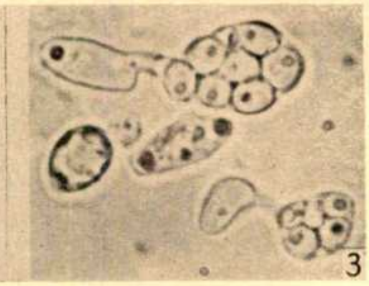


Fig. 1. *Pichia membranaefaciens*, strain *VB* 65 (*T* 109). Asci with bean-shaped spores. From *MA* 29 sporulation medium, after 5 days at 25° C. Unstained. (× 2,000)

Fig. 2. *Pichia membranaefaciens*, strain *Ac* 40. Asci with three and seven spores. From the pellicle formed in malt extract, after 3 days at 25° C. Unstained. (× 2,000)

Fig. 3. *Pichia membranaefaciens*, strain *Ac* 40. Asci with four and seven spores formed after heterogeneous conjugation. From the pellicle formed in malt extract, after 3 days at 25° C. Unstained. (× 2,000)

(*b*) From a sulphurized grape must which began to ferment spontaneously four months after preparation, among other yeasts a culture, *VB* 65 (*T* 109), was isolated, which was a characteristic strain of *Pichia membranaefaciens* but possessed the peculiarity that, as well as the normal spores, it also formed kidney-shaped spores; such spores have never been mentioned in the descriptions of the genus *Pichia*, not even in the very modern one by Phaff[4]. As a rule, great importance is given to the form of the spores as a taxonomic characteristic; for example, Kudriavtzev[2] created the genus *Fabospora* on the basis of its bean-shaped or reniform spores, and established a relation between this spore form and its weak ability for fermenting carbohydrates. Wickerham[7] also, on establishing his new genus *Dekkeromyces*, conceded special importance to the spore form. It appears very difficult to separate my organism from the species *Pichia membranaefaciens*, in which case the taxonomic value of the spore form will be open to controversy.

(*c*) From samples of 'alpechín' obtained from two different parts of the province of Badajoz, two yeast cultures were isolated, *Ac* 40 and *Ac* 47, which are two strains of *Pichia membranaefaciens*; although *Ac* 40 forms only a rather primitive pseudomycelium in anaerobiosis, the new definition of the genus *Pichia* given by Phaff[4] admits species without or with a reduced pseudomycelium. These two strains develop a smooth pellicle in malt after three days at 25° C. and form a large number of asci with 1–4 spores, as well as others with 5, 6 and 7 spores; asci are formed without immediately preceding conjugation or after heterogamous conjugation. These asci have not been mentioned in the genus *Pichia*, so that a revision of the diagnosis of *Pichia* is necessary.

(*d*) From different samples of 'alpechín' obtained from 22 places in different regions of Spain, 28 strains of *Saccharomyces oleaginosus*[5] were isolated. One of these strains, which has characteristics coinciding completely with those typical of the species, nevertheless differs from the remainder in its positive assimilation of potassium nitrate. According to Lodder and Kreger-van Rij[1] and Wickerham[6], yeasts which reproduce asexually exclusively by budding, form 1–4 ascospores per ascus and assimilate nitrate, should be included in the genus *Hansenula*. Nevertheless, in view of the observations mentioned, I doubt whether the assimilation of potassium nitrate can be regarded as a taxonomic characteristic of importance.

A detailed account of this work will be published elsewhere.

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[1] Lodder, J., and Kreger-van Rij, N. J. W., "The Yeasts: a Taxonomic Study" (North-Holland Pub. Co., Amsterdam, 1952).

[2] Kudriavtzev, V. I., "Sistematica Drozhzhey" (Academy of Sciences, Moscow, 1954).

[3] Santa María, J., Ann. Inst. Nac. Invest. Agronómicas (in the press).

[4] Phaff, H. J., *Ant. Leeuwenhoek. J. Mic. Serol.*, **22**, 113 (1956).

[5] Santa María, J., *Bol. Inst. Nac. Invest. Agronómicas*, **38**, 301 (1958).

[6] Wickerham, L. J., U.S. Dept. Agric. Tech. Bull. No. 1029 (1951).

[7] Wickerham, L. J., *J. Bact.*, **71**, 290 (1956).

Permeability of Dormant Bacterial Spores

THE frequent assumption that bacterial spores are impermeable and thereby metabolically dormant, unstainable and resistant to environmental stress has not heretofore been tested by experiment. We have attempted to examine this problem using clean, dormant, thermostable spores of a common aerobic bacterium, *Bacillus cereus* strain terminalis.

The organism was grown in a liquid semi-synthetic medium to essentially complete and synchronous sporulation[1]. After sporangial autolysis, the free spores were harvested by centrifugation or foam separation and washed repeatedly with distilled water to demonstrable removal of stainable cells and debris, catalase and germinating compounds. The total uptake of exogenous solutes by masses of the clean spores was assessed by the 'space' technique, the details and formulations of which have been described previously[2]. This procedure involves the addition of a known volume and concentration of solute to a known large mass of cells; after mixing and repacking the cells, the uptake is determined from the degree of dilution of the solute resulting from penetration into the cell pack. This uptake is expressed as a space value (S), which is defined as the fraction (or percentage) of the volume of the pack that the solute can penetrate. However, uptake into the cell pack involves space both between as well as inside the cells, and therefore correction for the interstitium must be made. With a non-penetrating macromolecule, such as a high molecular weight dextran, the intercellular space (S_{in}) for spores centrifuged at 17,000g for 30 min. was about 10 per cent but needed to be determined in each experiment. After appropriate allowance for this value, the uptake is expressed as a corrected space value (R), which is defined as the fraction (or percentage) of the volume of the cells themselves that the solute can penetrate.

Test of the impermeability hypothesis was made initially with glucose employed in a concentration of 10 mgm./ml. in distilled water at pH 6·8 and 0° C. with a 15-min. exposure period. After allowance for the intercellular space, the corrected mean glucose space (R) was found to be about 40 per cent whether measured isotopically, gravimetrically or by carbon combustion (Table 1). This uptake of glucose was essentially independent of temperature, pH, solute concentration, period of exposure and presence of other solutes or sodium azide and could be completely eluted with water. Thus, the observed uptake of glucose could not be accounted for by surface adsorption, exchange with internal glucose or metabolic removal. Rather, there appeared to be a net diffusional entry into a region occupying 40 per cent of the spore volume.

Table 1. UPTAKE OF GLUCOSE BY DORMANT SPORES

Analytical method	No. of experiments	R (%)		
		Range	Mean	*S.D.*
Gravimetric	8	31–47	40	1·4
Isotopic	4	34–51	40	5·7
Carbon combustion	7	34–47	42	3·9

If so, then other small molecules could be expected to permeate dormant spores to much the same extent. Indeed, a series of representative compounds that included organic acids, amino-acids, sugars and sugar alcohols gave space values approximating that for glucose (Table 2). Two types of compounds provided notable exceptions. Compounds such as adenosine gave variably high space values, and elution was possible only at the isoelectric point, suggesting some adsorptive uptake. Compounds such as ethylene glycol and urea gave constantly high space values (up to 57 per cent) which appeared to be real and possibly associated with lipid solubility. Murrell and Scott[3] have demonstrated that deuterium solution can mix freely with the total water volume of bacterial spores. We are now attempting to confirm, by means of the space technique, this seemingly complete permeation of water into the spore.

Table 2. UPTAKE OF VARIOUS SOLUTES BY DORMANT SPORES

Solute	R	Solute	R
Pyruvate	0·42	Glucose	0·40
Succinate	0·44	Ribose	0·41
Citrate	0·41	Maltose	0·44
DL-Alanine	0·32	Raffinose	0·42
DL-Valine	0·40	Erythritol	0·42
L-Lysine	0·42	Glycerol	0·44
L-Aspartate	0·41	Ethylene glycol	0·53
Adenine	1·10	Propylene glycol	0·53
Adenosine	0·76–1·2	Urea	0·57

Uptake of larger molecules was determined with a graded molecular weight series (62–2,000,000) of polyethylene glycols and dextrans. Space values for these two compounds were comparable and diminished linearly through a hundred-fold increase in molecular weight, suggesting that the surface structures of spores into which exogenous solute penetrates have the equivalent of pores which either vary in size or become progressively smaller with depth. A minimum uncorrected space value of about 10 per cent occurred with dextrans of molecular weight 100,000 or greater; this constant value was concluded to represent the intercellular space.

Concurrent and previous studies[1] on structural differentiation and associated physiological changes have intimated that diffusional uptake is limited to the multilayered thick coat of the spore. Two internal membranes, the cortex and the contiguous inner membrane, appear to be formed at the same time that the core, apparently, of a maturing spore becomes light-refractile and resistant to stains. A mature spore with a fully formed cortex actually takes up stains, as measured with the space technique, and can be seen to absorb stain in its peripheral structure. Furthermore, such a spore can be totally stained after it has been crushed, indicating that the core is not chromophobic but that a barrier shielding it has been broken. Attempts also were made to localize the observed uptake by comparing average R values with direct measurements of spore-coat space. Calculations from measurements on electron micrographs of medial ultra-thin sections indicated that the volume occupied by spore membranes external to the core is approximately 40 per cent of the total spore volume, assuming spores to be ellipsoids of revolution and the spore structure to be of uniform density. This apparent localization of solute permeation in the peripheral membranes of dormant spores is being investigated further.

Detailed reports of these findings will be published elsewhere.

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[1] Hashimoto, T., Black, S. H., and Gerhardt, P., *Canad. J. Microbiol.* (in the press).

[2] MacDonald, R. E., and Gerhardt, P., *Canad. J. Microbiol.*, **4**, 109 (1958). See also Britt, E. M., and Gerhardt, P., *J. Bacteriol.*, **76**, 288 (1958).

[3] Murrell, W. G., and Scott, W. J., Seventh Int. Congr. Microbiol., Absts., 26 (1958).

Decontaminating Tissue Cultures infected with Pleuropneumonia-like Organisms

THE contamination of tissue culture cell lines by pleuropneumonia-like organisms has become an increasingly important problem[1–4]. The contamination itself is insidious and cannot be detected ordinarily by gross changes in the appearance of the media macroscopically or microscopically. The tissue culture cells themselves, when observed microscopically, may or may not exhibit slight cytopathology as evidenced by intracytoplasmic granularity. Properly stained preparations may reveal intracytoplasmic particles, presumably the contaminating pleuropneumonia-like organisms[5–7].

At present it is not understood how these organisms gain entrance to cultures, although speculation that they represent *L* forms of bacterial contaminants seems to provide the most likely explanation[4]. Most pleuropneumonia-like organisms are resistant to penicillin but susceptible in varying degrees to streptomycin and certain of the broad-spectrum antibiotics[8,9]. Certain evidence indicates that the presence of high concentrations of antibiotics to which pleuropneumonia-like organisms are sensitive drives these organisms into intracellular residence, and that when the antibiotic concentration drops below a critical level (due to long incubation at 37° C. or extremes of *p*H) the organisms are free to reappear in the supernatant fluids. This cycle of appearance and disappearance of pleuropneumonia-like organisms in tissue culture fluids containing antibiotics must be considered when isolations of organisms are attempted from tissue cultures.

Since routine sampling of tissue culture media components including serum has not as yet revealed pleuropneumonia-like organisms, another explanation for the source of contamination must be considered. The suggestion that cell lines have been contaminated from the tissue of origin is probably not valid since: (1) routine sampling of primary tissue cultures has not revealed these organisms and (2) studies of HeLa cell cultures have not substantiated interpretations favourable to the hypothesis. The widespread prevalence of pleuropneumonia-like organism contamination of HeLa cell cultures and the frequent occurrence of pleuropneumonia-like organisms in the urogenital tract had been regarded as evidence that contamination occurred when the cell line was first isolated. However, routine testing of a number of HeLa sublines in 1953, when HeLa was first generally available, always gave negative results[5]. The techniques used in the attempted isolations at that time were the same as those used recently which resulted in positive isolations[4].

Accordingly, another explanation has been offered, that the pleuropneumonia-like organisms probably represent *L* forms of bacterial contaminants[4]. A complete list of the kinds of environmental trauma that can induce bacteria to revert to *L* forms is lacking. It is known, however, that agents like glycine, raffinose, salts of light metals, carboxymethoxylamine, chloromycetin, cold, X-rays, phage, glycerol and most of all penicillin, can cause this reversion[10]. This list includes a number of reagents that are used routinely in the tissue culture technique. Therefore the hypothesis is proposed that the *L* forms present in tissue cultures were induced by the action of penicillin, and also perhaps by glycine, salts of light metals, and glycerol, on bacteria. This process occurs when breaks in aseptic technique allow for the presence of contaminating bacteria which, under the influence of one of these reagents, revert to the corresponding *L* form.

A method for decontaminating pleuropneumonia-like organism (or *L* form) contaminated tissue cultures has been developed and is based on the differential heat lability of the pleuropneumonia-like organisms and the tissue culture cells. It has been found that if tissue culture cell lines, such as HeLa, *L* cells, *WISH* (a human amnion line), Fernandes amnion, and a human renal line, have been either naturally or artificially contaminated with pleuropneumonia-like organisms and are then held at 41° C. for about 18 hr., they remain negative for pleuropneumonia-like organisms indefinitely. The pleuropneumonia-like organisms are very heat-labile. Some strains are killed at 45° C. for 15 min., others succumb at 45° C. for 30 min.[11]. Tissue culture cells held at 45° C., in general, do not survive, yet when incubated at 41° C. for a long period of time differential survival is achieved—the pleuropneumonia-like organisms are killed and the tissue culture cells survive.

This hyperthermic treatment of cell lines is not without some effect on the cells, and tolerance to 41° C. varies from cell line to cell line, although they all apparently will survive[12]. HeLa at 41° C. has a tendency toward arborization, withdrawal of processes, some rounding and cytoplasmic vacuolization. However, this is by no means fatal to the culture, and once the cells are placed at 37° C. after treatment, recovery is almost always assured. Confirmation of the heat lability of two tissue culture pleuropneumonia-like organism contaminants was obtained when these strains were found to be killed when grown out on agar and then subjected to 41° C. for 18 hr. It is recommended that for optimum simplicity the heat treatment be done in air, since if a water-bath is used it will necessitate complete submersion of the tissue culture vessel in water, with concomitant problems of possible water leakage through the stopper.

Evidence has recently been offered that aureomycin in doses toxic to the tissue culture cells was successful in eliminating contamination by pleuropneumonia-like organisms[13]. This technique entails the use of an antibiotic not normally used in tissue culture, and the material must be added at each media change every 2 or 3 days. Additionally, it is known that there is a great variability in the toxic levels of aureomycin for any particular tissue culture cell line[13]. This would necessitate experimentation with each tissue culture cell line to determine the toxic dose. The hyperthermic technique obviates this disadvantage.

The advantages of the hyperthermic technique are: (1) no new chemical or antibiotic is introduced into the culture; (2) there is assurance that the pleuropneumonia-like organisms are killed, not merely arrested in growth by the bacteriostatic effect of antibiotics[11]; (3) intracellular pleuropneumonia-like organisms are also killed; (4) the method is very simple and easy to carry out.

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[1] Robinson, L. R., Wichelhausen, R. H., and Roizman, B., *Science*, **124**, 1147 (1956).

[2] Collier, L. H., *Nature*, **180**, 757 (1957).

[3] Rothblatt, G. O., and Morton, H. E., *Bact. Proc.*, **73** (1958).

[4] Hayflick, L., and Stinebring, W. R., paper presented at a Conference on the Biology of the Pleuropneumonia-like Organisms, *Ann. N.Y. Acad. Sci.*, **79**, 433 (1960).

[5] Hayflick, L., and Stinebring, W. R., *Anat. Rec.*, **121**, 477 (1955).

[6] Hayflick, L., Stinebring, W. R., Breckenridge, F. C., and Pomerat, C. M., *Bact. Proc.*, **75** (1956).

[7] Shepard, M. C., *J. Bact.*, **75**, 351 (1958).

[8] Leberman, P. R., Smith, P. F., and Morton, H. E., *J. Urol.*, **64**, 167 (1950).

[9] Leberman, P. R., Smith, P. F., and Morton, H. E., *J. Urol.*, **68**, 399 (1952).

[10] Tulasne, R., *Revue d'Immunologie*, **15**, 223 (1951).

[11] Edward, D. G. ff., *J. Gen. Microbiol.*, **10**, 27 (1954).

[12] Selawry, O. S., Goldstein, M. N., and McCormick, T., *Cancer Res.*, **17**, 785 (1957).

[13] Hearn, jun., J. H., Officer, J. E., Elsner, V., and Brown, A., *J. Bact.*, **78**, 574 (1959).

PATHOLOGY

Cerebellar and Midbrain Lesions in Scrapie

ATAXIA, inco-ordination of movement and to a limited extent disturbances in muscular tonus appear to be the main neurological symptoms in scrapie disease of sheep. Such symptoms suggested cerebellar lesions and indeed lesions have been demonstrated in the cerebellum[1-3], but only occasionally. Recently, an alteration has been made in our technique of taking material from the cerebellum for the routine histological examinations. Dorso-ventral sections have been taken through the middle of the cerebellum, parallel to the long axis of the brain, instead of the usual slice from a sagittal section through the vermis. Examination of such sections from nine brains of sheep affected with natural scrapie showed in each case degeneration and vacuolation of the Purkinje cells in a number of the folia of the paraflocculus (Fig. 1), while occasionally the neurones of the dentate nucleus were also affected. In the midbrains of these animals, in addition to vacuolation of the pigmented cells of the raphe, chromatolysis of the neurones of the red nucleus was a constant finding, while vacuolation was also often present. In the pons the neurones of the nucleus of the abducens nerve were always degenerated and vacuolated, but in the cells of the reticular formation changes were present in seven cases out of nine. In the medulla, lesions were found in the nuclei of the raphe, arcuate and formatio reticularis alba. Lesions of the above nuclei of the medulla have previously been shown to be constantly present in both natural and experimental scrapie[3,4].

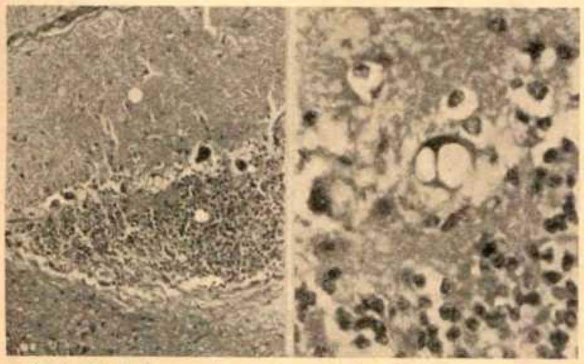

Fig. 1. Left, degeneration and vacuolation of the Purkinje cells (× 90); right, high-power magnification of a vacuolated Purkinje cell (× 360)

In the experimentally induced disease, although the clinical symptoms are identical with those of natural scrapie, lesions in the medulla and pons are not always very obvious[4] and although the distribution of such lesions as there are is similar to that seen in the naturally occurring disease the neurones undergoing degeneration are occasionally so few in number that it is difficult to relate the symptoms to the lesions. In the majority of experimental scrapie cases so far examined the neurones of the red nucleus of the mesencephalon were often affected as also were cells in the medulla and pons, but on occasion no lesions could be found in the midbrain even when 18 consecutive serial sections were examined. Recently, however, transverse sections of the mesencephalon from three such brains have been examined by cutting serial sections along the whole of the red nucleus. A total of 1,000 sections 7μ thick were cut but only every fourth section was examined, and in each of the three midbrains a varying number of the large neurones of the red nucleus was found undergoing either chromatolysis, pyknosis or necrosis. It would seem, therefore, that in all cases of scrapie the neurones of the red nucleus are affected. It is known that in lower mammals including the sheep the red nucleus serves as a centre through which the cerebellum can influence the motor functions of the spinal cord and medulla oblongata[5]. Lesions in the red nucleus and cerebellum will therefore cause lack of co-ordination of muscular activity as well as disturbances in tonus and synergy.

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Gilmerton, Edinburgh. Nov. 11.

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[1] Bertrand, I., Carré, H., and Lucam, F., *Rec. Méd. vét.*, **113**, 586 (1937).

[2] Wagner, A. R., Goldstein, H. E., Doran, J. E., and Hay, J. R., *J. Amer. Vet. Med. Assoc.*, **124**, 136 (1954).

[3] Zlotnik, I., *J. Comp. Path.*, **68**, 148 (1958).

[4] Zlotnik, I., *J. Comp. Path.*, **68**, 428 (1958).

[5] Ranson, S. W., "The Anatomy of the Nervous System" (W. B. Saunders Co., 1953).

Effect of Injection of a Hepatotoxin directly into the Gall-Bladder of Rabbits

AN inflammatory condition of the bile ducts is generally recognized to be the essential feature of 'facial eczema' liver damage in sheep[1]. This condition has recently been shown to be caused by a toxic principle elaborated by the fungus *Sporidesmium bakeri* Syd., a common species inhabiting many pastures in New Zealand (ref. 2 and Percival, J. C., unpublished work). Guinea pigs and rabbits have also been shown to be susceptible to the disease, liver damage of a generally similar nature having been produced by feeding toxic grass and crude fungal culture, and ether-extracts of these (Clare, N. T., unpublished work), and also by injection of extracts (Worker, N. A., unpublished work).

Recently, in a series of experiments on rabbits, in which the relative effectiveness of various routes of administration was compared, ether extracts of fungus, dispersed in 5 per cent aqueous 'Tween 40', were injected into: (1) the duodenum, proximal to the sphincter of Oddi; (2) the portal vein; (3) the gall-bladder.

Twenty New Zealand white rabbits of mixed sex, weighing 1,000–1,500 gm., were allotted to each group at random. Each rabbit was dosed with the equivalent of 1 litre of fungal culture dispersed in 0·4 ml. 'Tween'. The animals were killed at periods varying from four to seven days. Examination of the morbid anatomy and histology of the livers was carried out by D. C. Dodd, veterinary diagnostic officer of this Station.

From the results summarized in Table 1, it can be seen that after injection of extracts into the duodenum and into the portal vein, the majority of animals in both treatment groups exhibited mild to moderate liver damage, the lesions being similar both macroscopically and histologically to those commonly found after administration of toxic material by mouth (Dodd, D. C., unpublished work). In contrast to these results, however, injection of toxic extract into the gall-bladder produced, in every animal, lesions of the most severe form. The end result of this direct injection procedure was inevitably functional obliteration of the gall-bladder, with very severe generalized

Table 1. GRADINGS OF LIVER LESIONS IN RABBITS AFTER ADMINISTRATION OF FACIAL ECZEMA TOXIN BY DIFFERENT ROUTES
Gradings based on macroscopic and histological examination

Route of administration	Grading			
	Mild	Moderate	Severe	Very severe
(1) Duodenum	7	11	2	0
(2) Portal vein	6	10	4	0
(3) Gall-bladder	0	0	0	20

œdema and fibrosis of the intra- and extra-hepatic biliary systems, together with gross enlargement of the liver and marked discoloration. A most outstanding feature of the results was the completely generalized effects of the treatment throughout all lobes of the liver. Twelve of the twenty animals so treated exhibited clinical icterus, a condition which was not noted in either of the other experimental groups and which in the past has been seldom encountered in rabbits in the course of other studies relating to this disease.

The results of the above studies represent a somewhat perplexing situation. In particular, the completely generalized effect noted throughout the liver of every animal in group 3 is puzzling and is difficult to explain satisfactorily in terms of the accepted facts of liver physiology and function. Expulsion of the toxic dose by way of the common bile duct followed by its subsequent resorption through the intestine would appear to be ruled out as a major contributing factor in view of the finding that injection of equivalent amounts of toxin into the duodenum (treatment 1) or the portal vein (treatment 2) produced damage of very much lesser severity. Similarly, it would appear that absorption of the dose through the venous drainage of the gall-bladder is not a factor of any great significance since cystic venous blood discharges into the portal circulation immediately adjacent to the liver, a situation as represented in treatment 2.

One possible explanation for this effect might be a retrograde movement of toxin from the gall-bladder into the biliary tree. Although it is difficult to visualize a diffusion of toxin against a bile flow which has been shown in this laboratory, in animals of similar size, to be of the order of 6–10 ml. per hr., such a hypothesis would provide an adequate explanation for the extreme severity of the lesions and for the uniform distribution of damaged biliary epithelial tissue throughout the entire substance of the liver.

This hypothesis clearly requires further investigation, not only in view of its general interest, but more particularly because of its possible implications in the pathogenesis of liver damage in facial eczema. If it can be demonstrated that, after concentration and storage in the gall-bladder, materials can re-enter and move freely within the biliary tree, it would suggest that in animals grazing under field conditions the function of the gall-bladder and biliary apparatus may be an important factor in the development of liver lesions. Attempts are being made with the aid of radio-opaque tracers to study gall-bladder function in more detail and to follow more closely the course taken by materials in transit from the gall-bladder to the intestine.

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[1] Cunningham, I. J., Hopkirk, C. S. M., and Filmer, J. F., *N.Z. J. Sci. Tech.*, **24A**, 185 (1942).

[2] Thornton, R. H., and Percival, J. C., *Nature*, **183**, 63 (1959).

Effects of Dehydration on the Mitochondria of the Mouse Kidney

THE effect of complete withdrawal of water on the animal organism has been studied very little from the point of view of morbid anatomy. The changes observed have been included in the signs of general cell damage such as cloudy swelling, for example.

Modern histochemical techniques[1,2] have made it possible to get information not only about intracellular oxidative mechanisms but also about the physical state of the mitochondria. Employment of the techniques in this way can be referred to as 'mitochondrial assay'[3]. It has been possible to localize cellular damage within a few hours in sites where lesions observable with the classical methods of histology develop only after some days or weeks. By this means it has been shown that swelling of the mitochondria of the kidney is a very early sign of pathological change in a number of conditions, such as magnesium depletion[4], starvation[5], and vitamin D overdosage[6].

The effect of complete dehydration on the mouse kidney has now been studied using histochemical reactions for succinic dehydrogenase and diphosphopyridine nucleotide diaphorase[7]. Mice were killed after 1, 2, 3 and 4 days complete deprivation of water.

With both dehydrogenase and diaphorase methods mitochondrial swelling could be observed after only one day of water deprivation. The intramitochondrial formazan deposits became larger in a limited number of tubular cells, distributed over the whole nephron. During subsequent days mitochondrial swelling increased progressively; in Fig. 1 is shown the reaction for diphosphopyridine nucleotide diaphorase after 4 days of water deprivation. When the animals were allowed to drink water after 4 days dehydration, the mitochondrial changes were rapidly reversed and the picture returned to normal, as shown in Fig. 2.

It is known that the laboratory animals can concentrate their urine to a much greater extent than

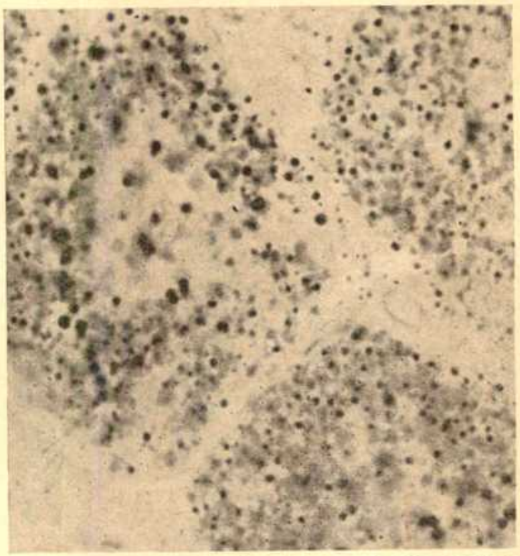

Fig. 1. Reduced diphosphopyridine nucleotide diaphorase reaction of the mouse kidney after four days complete water deprivation (× 1,800)

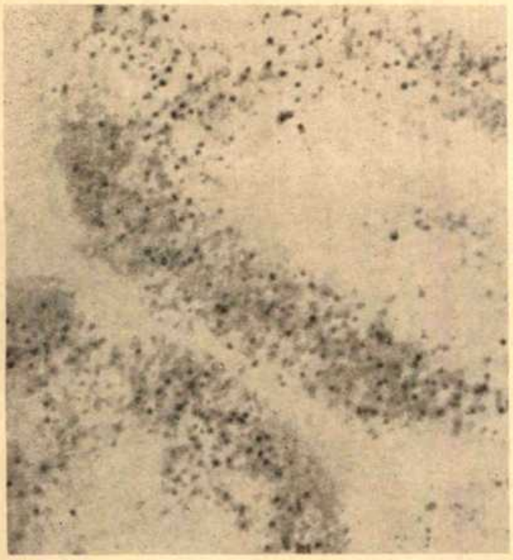

Fig. 2. Same reaction 24 hr. after allowing the dehydrated animal free access to water (× 1,800)

man. It has been believed that this ability to concentrate is a physiological function causing no morphological change in the cells. The results presented here suggest, however, that the high concentration of urine is achieved at the expense of a reversible mitochondrial damage.

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[1] Pearse, A. G. E., and Scarpelli, D. G., *Nature*, **181**, 702 (1958).
[2] Hess, R., Scarpelli, D. G., and Pearse, A. G. E., *Nature*, **181**, 1531 (1958).
[3] Pearse, A. G. E., *J. Clin. Path.*, **11**, 520 (1958). "Histochemistry, Theoretical and Applied", second ed. (J. and A. Churchill, London, 1960).
[4] Hess, R., Macintyre, I., Alcock, N., and Pearse, A. G. E., *Brit. J. Exp. Path.*, **15**, 80 (1959).
[5] Scarpelli, D. G., *Exp. Cell. Res.* (in the press).
[6] Scarpelli, D. G., Tremblay, G., and Pearse, A. G. E., *Amer. J. Path.* (in the press).
[7] Scarpelli, D. G., Hess, R., and Pearse, A. G. E., *J. Biophys. Biochem. Cytol.*, **4**, 747 (1958).

A New Approach to Tumour Immunity

ONLY rarely and depending on special conditions has actively acquired immunity been demonstrated for tumours indigenous to and tested within inbred strains of mice[1-3] or rats[4]. For this reason doubts have been expressed as to whether 'tumour immunity' exists independent of isoantigenic differences. Experiments carried out in this laboratory and summarized in Table 1 have utilized inoculation into mice of inbred strains of tumours produced in F_1 hybrid mice derived from the corresponding parent strains. Mice of the parent strain in which the F_1 'hybrid tumour' failed to grow were challenged with a tumour indigenous to and specific for their own strain.

The parent strains were *C57BL*, *C3H*, and *AKR*, from which F_1 hybrids (reciprocal crosses) *C57BL* × *AKR* and *C57BL* × *C3H* were produced. Tumours induced in F_1 hybrids by subcutaneous injection of methylcholanthrene were serially transplanted into F_1 mice of the same origin. Tumour *CAF* in (*C57BL* × *AKR*) F_1 was an infiltrating fibrosarcoma, tumour *CCF* in (*C3H* × *C57BL*) F_1 a moderately well-differentiated mammary carcinoma; both tumours grew in 100 per cent of inoculated homologous F_1 mice. When inoculated into either of the parent strains, these hybrid tumours grew in some animals if sufficiently large numbers of tumour cells were given. The ability of F_1 tumours to grow in parent strains, in spite of immunogenetic differences, has been thoroughly analysed by Klein *et al.*[5]. We have as yet not investigated in detail factors affecting the transplantability of hybrid tumours *CCF* and *CAF* to mice of parent strains. After inoculation of hybrid tumours in increasing doses a percentage of mice of the three parent strains remained free of tumour growth. Upon challenge of *C57BL* mice with sarcoma-89, a fibrosarcoma indigenous to and specific for this strain[6], most of these animals proved resistant to this tumour, as shown in Table 1, while control *C57BL* mice inoculated at the same time succumbed to progressive tumour growth. No such immunity was observed when *C3H* and *AKR* mice, respectively, following inoculation of F_1 hybrid tumours, were afterwards subjected to challenge with tumour indigenous to their strain (transplantable *C3H* mammary carcinoma; *CAM* leukæmia of *AKR* strain): killing tumours developed in 100 per cent of these mice as they did in control mice.

At present no explanation can be offered for the immunizing ability of F_1 hybrid tumours for *C57BL* mice but not for *C3H* and *AKR* mice. Two possibilities may be considered: (1) specific immuno-

Table 1. IMMUNITY TO STRAIN-SPECIFIC TUMOUR OF *C57BL* MICE INOCULATED WITH F_1 TUMOUR

Experiment No.	No. of mice		F_1 tumour inoculation*				No. of mice resistant to F_1 tumour		Sarcoma-89 inoculation*		No. of mice resistant to sarcoma-89	
	M	F	Type†	I	II	III	M	F	I	II	M	F
1	5	5	*CAF*	$2 \cdot 5 \times 10^5$	$3 \cdot 5 \times 10^5$ (8)	—	0	3	3×10^5 (6)	3×10^5 (9)	0	3
2	6	6	*CAF*	1×10^5	$3 \cdot 5 \times 10^5$ (4)	5×10^5 (3)	6	6	3×10^5 (4)	—	6	6
3	5	5	*CCF*	2×10^5	$5 \cdot 5 \times 10^5$ (3)	7×10^5 (3)	5	5	$3 \cdot 4 \times 10^5$ (4)	—	5	5
4	20	30	*CAF*	1×10^5	5×10^5 (3)	—	4	26	4×10^5 (5)	—	3	15

* Figures indicate number of viable (unstained) cells injected per mouse, with parenthetical numbers representing time interval (weeks) since preceding inoculation.

† For explanation, see text.

genetic mechanisms may be involved, such as presence of a specific antigen common to F_1 hybrid tumour and sarcoma-89. In this connexion, it should be noted that while the *CAF* tumour was derived from a methylcholanthrene-induced sarcoma as was also sarcoma-89, the *CAF* hybrid tumour originated from a mammary carcinoma of a host injected with methylcholanthrene. Study of possible immunizing ability present in normal tissues of F_1 has been initiated by injecting *C57BL* mice with splenic cells of (*C57BL* × *AKR*) F_1 mice prior to inoculation with sarcoma-89. (2) Another possibility is suggested by previous studies in which we found *C57BL* mice to be much more potent antibody producers than *C3H* and *AKR* mice, as judged from levels of antibodies after injection of heterologous red cells[7]. On this basis one might suspect that only *C57BL* mice, but not *AKR* and *C3H* mice, produced sufficient antibody to F_1 hybrid tumours.

This work is now being expanded, with inclusion of additional systems testing immunizing ability of F_1 tumours in other inbred strains in order to elucidate the phenomenon described.

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[1] Foley, E. J., *Proc. Soc. Exp. Biol. and Med.*, **80**, 675 (1952).

[2] Prehn, R. T., and Main, J. M., *J. Nat. Cancer Inst.*, **18**, 769 (1957).

[3] Prince, J. E., Fardon, J. C., Nutini, L. G., and Sperti, G. S., *Cancer Res.*, **17**, 312 (1957).

[4] Goldner, H., Bogden, A. E., and Aptekman, P. M., *J. Immunol.*, **12**, 520 (1959).

[5] Klein, E., and Klein, G., *Transplant. Bull.*, **3**, 136 (1956).

[6] Dunham, L. J., and Stewart, H. L., *J. Nat. Cancer Inst.*, **13**, 1299 (1953).

[7] Davidsohn, I., and Stern, K., *J. Immunol.*, **72**, 216 (1954).

Presence of 3-Methoxy-4-hydroxyphenylglycol and Metanephrine in Phæochromocytoma Tissue

THE ethereal sulphate of 3-methoxy-4-hydroxyphenylglycol, a metabolite of epinephrine and norepinephrine, has recently been demonstrated in the urine of normal individuals and was noted to be markedly increased in the urine of patients having phæochromocytoma[1]. The free compound has not been previously demonstrated and the possibility that conjugated (nor)metanephrine gives rise to this compound could not be ruled out. We have now been able to demonstrate the presence of free 3-methoxy-4-hydroxyphenylglycol in phæochromocytoma tissue from four different patients, as well as metanephrine in three of these tumours.

About 5 gm. of phæochromocytoma tissue was homogenized with 2 vol. of 0·5 *M* acetate buffer at *p*H 3, saturated with sodium chloride and extracted into 10 vol. of ethyl acetate. The ethyl acetate extract was then washed with salt-saturated acetate buffer at *p*H 7 and evaporated to dryness *in vacuo*. The residue was taken up in 10 ml. acetone, transferred to a 15-ml. centrifuge tube, and the solvent evaporated in a current of nitrogen until the volume was about 4 c.c. After centrifugation, 0·5 ml. of the supernatant fluid was applied to Whatman No. 1 paper and subjected to two-dimensional chromatography using *iso*propanol/5 per cent ammonia (8 : 2) and *n*-butanol/acetic acid/water (8 : 8 : 2) as the solvent systems. A spot was present having the same R_F and colour reactions as authentic 3-methoxy-4-hydroxyphenylglycol[1]. Addition of the authentic substance to the chromatogram increased the intensity of this spot. From the intensity of the colour of the spot, it was estimated that 5–10 μgm. of the glycol per gm. of tissue was present in the tumours.

Another spot, metanephrine[2], was identified in a similar manner and was shown to be different from normetanephrine by separation during chromatography. This substance was demonstrated in three of the four tumours.

The demonstration of unconjugated 3-methoxy-4-hydroxyphenylglycol of biological origin has not been previously reported. This supports the pathway for formation of the sulphate of this metabolite from the free compound, rather than from (nor)metanephrine sulphate. Although normetanephrine has been reported in phæochromocytoma[3], this is the first demonstration of the presence of metanephrine in these tumours.

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[1] Axelrod, J., Kopin, I. J., and Mann, J. D., *Biochim. Biophys. Acta* (in the press).

[2] Axelrod, J., Senoh, S., and Witkop, B., *J. Biol. Chem.*, **233**, 697 (1958).

[3] Sjoerdsma, A., *et al.*, *Science*, **127**, 876 (1958).

A Soluble Protective Antigen of *Trypanosoma brucei*

Trypanosoma brucei in quantities sufficient for antigenic analysis was obtained by harvesting the blood of rats on the third day of infection by the protozoon, when the fulminating parasitæmia is at its height. The trypanosomes were separated from the blood by centrifugation and the resulting mash was preserved by freeze drying. Several batches made in this way were pooled.

Rabbits inoculated with this material produced not only agglutinins for live trypanosomes but also antibodies precipitating with rat serum. These precipitins, a response to traces of serum proteins not removed during the separation of the trypanosomes from the blood, were removed by absorption with normal rat serum. The absorbed antiserum, no longer reacting with normal rat serum, nevertheless precipitated with some component in the serum of infected rats, from which the trypanosomes had been removed by filtration ; suggesting that infected rat serum contained an antigen peculiar to the infected state. This antigen was designated the 'exoantigen' of *T. brucei*.

Antisera to the exoantigen, free of antibodies to normal rat serum, were prepared by inoculating normal rats with trypanosome-free infected rat serum. These antisera not only precipitated exoantigens, but also agglutinated living trypanosomes *in vitro*, suggesting strongly that the exoantigens were liberated from trypanosomes and were not of rat origin. About a million living trypanosomes, which had been washed as thoroughly as possible, were injected into rabbits and in two or three weeks the protozoa proliferated sufficiently to produce the typical chronic infection, and to induce antibodies, which agglutinated trypanosomes and precipitated the exoantigen present in infected rat serum. It is evident from quantitative considerations that the precipitins were not induced by an abnormal rat metabolite carried on the original inoculum but by antigens liberated from the multiplying trypanosomes. First, the amount of rat serum proteins contaminating the inoculum was insufficient to stimulate antibodies to normal serum proteins and therefore to the certainly much smaller amounts of exoantigen present. Secondly, the million trypanosomes injected could not have been carrying an antigenically effective amount of an abnormal rat metabolite because grams of wet weight of dead trypanosomes from the rat are needed to induce a similar antibody response in rabbits.

The *in vitro* agglutination of living trypanosomes both by rabbit antisera to living trypanosomes and by rabbit or rat antisera to exoantigen was specifically inhibited by exoantigen. In gel-diffusion precipitin systems, the exoantigen formed a continuous line when opposed to adjacent reservoirs of each of the two kinds of antisera. Thus, antibody of the same specificity was induced whether the provoking antigen was living and multiplying trypanosomes or exoantigens in infected rat serum free of trypanosomes, leaving no doubt that the exoantigen was liberated from the trypanosomes and was not an abnormal rat metabolite. In an immuno-electrophoretic analysis of infected rat serum exoantigen appears as a slow-moving component, moving even more slowly than the serum γ-globulin.

Certain biological effects of free exoantigen indicate that it is concerned with the process of infection. When trypanosomes are collected from infected animals their viability *in vitro* largely depends on the type of medium in which they are contained. Washed suspensions of living trypanosomes were re-suspended in (1) Alsever's solution, (2) Alsever's solution plus normal rat serum, and (3) Alsever's solution plus infected rat serum containing exoantigen; they were injected into mice after standing at room temperature for 1–2 hr. Suspensions in Alsever's solution alone were usually non-infective. The time to death after injection of serum-treated preparations depended not only on the dose of trypanosomes, but also on the nature and amount of the added serum. Mice receiving trypanosomes and infected rat serum invariably died first, and the time to death was directly proportional to the concentration of exoantigen added. Mice receiving trypanosomes and normal rat serum always died later, if at all. Moreover, when suspensions of trypanosomes in normal and infected rat serum were held for varying periods on the bench, the older suspensions were the less infective, although the suspensions containing exoantigen remained infective for longer periods than those with normal rat serum. Since exoantigen appears to be relatively stable, it is unlikely to have been acting as an aggressin the potency of which was declining during the period on the bench; so that the increased infectivity of suspensions containing exoantigen appears mainly to be due to the more efficient preservation of the viability of the trypanosomes *in vitro*.

Nevertheless, though the exoantigen does not in these circumstances behave as a soluble aggressin, it has immunogenic properties. Repeated injection of exoantigen into mice and rats actively immunizes the animals against infection by live trypanosomes. Presumably, therefore, antibody to exoantigen is protective.

In conclusion, the exoantigen of *T. brucei* which occurs in the serum of infected rats plays an essential part in infection and is a means by which laboratory animals can be immunized against *T. brucei* by syringe inoculation. Its immunogenic efficiency against the infection transmitted by the tsetse fly—an experimental system closer to the natural disease—is under investigation.

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Runt Disease and Tolerance in Rabbits

The production of immunological tolerance, that is, specific inhibition of the immune response in adult life by contact with antigen at an early age, was first described by Billingham, Brent and Medawar[1] and Hašek[2]. By this means, animals of different strains or breeds can be made to accept skin homografts from each other in adult life, provided they have been injected in the fœtal or early neonatal period with cells taken from their future skin donors.

The time in early life (adaptive period) during which the animal reacts to the administration of foreign cells by becoming tolerant varies considerably from species to species. In rats[3] it ends some days after birth, in mice at birth[4], but in rabbits incomplete evidence suggests it probably ends some time before the end of intra-uterine existence[5]. Moreover, when the cells used to induce this tolerance are adult and capable of forming antibody, a considerable proportion of the recipients may develop runt disease, characterized by chronic wasting and premature death[4].

Recently, as part of a study of graft-against-host reactions in the rabbit, it became necessary to produce runts in this species. As this has not been described before, and as detailed information on the adaptive period in rabbits was incidentally obtained, the results may be of interest to others.

Homologous spleen cells from adult female chinchilla rabbits were injected into newborn or fœtal rabbits. The donor animal was anæsthetized and splenectomy performed. The spleen was finely chopped in heparinized saline, and then suspended by gently drawing up and down through a wide-bore cannula. After filtration through nylon bolting cloth of 90-μ porosity, a cell count was made and a suitable quantity of the spleen suspension injected into the recipient. Newborn rabbits were injected intravenously within 1–4 hr. of birth. To inject fœtuses, a laparotomy was performed on a pregnant rabbit and the uterus delivered through the wound. The head and back of each fœtus were identified, and then spleen suspension injected into the peritoneal cavity of each. The uterus was returned to the abdomen and the wound sutured. When possible, the recipients of spleen cell suspension were challenged at 8 weeks with skin grafts from the spleen donor, skin homografts from another unrelated donor and autografts. These were full-thickness grafts 2 cm. in diameter sutured into prepared beds upon the ears[6].

Table 1. Incidence of Runt Disease and Survival Time of Skin Homografts in Rabbits given Homologous Spleen Cells in Fœtal Life or at Birth

Group	No. of rabbits injected	Route of injection	Age at time of injection	No. of dead at birth	No. of runts	Survival times of skin grafts from spleen donors (days)
25 × 10^6 cells injected						
I	8	Intravenously	1–4 hr. after birth	0	0	6, 6, 6, 6, 7, 7, 8, 9
II	10	Intraperitoneally	28th day in utero	1	0	6, 6, 6, 7, 7, 8, 8, 8, 9
III	9	,,	26th day in utero	1	0	6, 6, 7, 7, 7, 8, 8, 9
IV	10	,,	24th day in utero	1	0	9, 9, 9, 9, 10, 10, 11, 11, 12
V	12	,,	22nd day in utero	2	1	7, 8, 9, 12, 18*, 20, 24, 28, 34, 180
VI	12	,,	20th day in utero	5	3	14*, 24*, 36, 40, 80, 180
50 × 10^6 cells injected						
VII	18	,,	22nd day in utero	7	5	7, 8, 16*, 24, 38, 180
VIII	21	,,	20th day in utero	9	8	14*, 68, 90, 180, 180

* Runts with viable skin grafts at time of death of host.

It can be seen from Table 1 that the adaptive period in the rabbit ends about the twenty-second day of gestation (normally pregnancy lasts 31 days). Giving rabbits 25 (× 10^6) spleen cells intravenously immediately after birth or intraperitoneally any time after the twenty-second day of fœtal life failed to produce immunological tolerance or runt disease.

Indeed, in fœtuses injected at the twenty-sixth day of pregnancy or later, skin homografts from the spleen donor were rejected more rapidly than corresponding homografts from unrelated rabbits, suggesting that immunity had resulted and that these pieces of skin were behaving as 'second-set' grafts[7]. In the 25 animals from groups I, II and III that were skin grafted, the mean survival times for the 'spleen donor' and 'unrelated' skin transplants were found to be $7\cdot12 \pm 1\cdot05$ and $10\cdot04 \pm 1\cdot21$ days, respectively, a difference which is significant ($P < 0\cdot01$).

After the critical twenty-second day there seems to be a brief 'neutral' period in which foreign cells are unable to induce either tolerance or immunity. Thus, in animals exposed to spleen cells at the twenty-fourth day of fœtal life (Group IV), there is no significant difference ($P > 0\cdot10$) between the survival times of skin homografts from the spleen donors and those of grafts from unrelated donors.

Of the 12 fœtal rabbits injected at 22 days (Group V), 6 became tolerant and showed definite prolongation of survival of skin homografts from the spleen donor, but not from the unrelated donor. Tolerance was not complete, and only one animal has an intact graft at the time of reporting (180 days). In another of the animals runt disease was encountered, as was only to be expected after the introduction of adult immunologically competent cells into an immature animal, and one of the two animals dead at birth showed the lymphoid atrophy usually associated with this syndrome.

In Group VI (injected at 20 days) all the surviving animals showed tolerance of varying degrees to spleen donor skin grafts. Runt disease was more common than in Group V and three of the five animals born dead showed evidence of the same process.

Increasing the dose of spleen suspension injected into 22- and 20-day old fœtuses to 50 (× 10^6) nucleated cells greatly raised the incidence of runt disease and the number of rabbits born dead (Groups VII and VIII). 9 of the 16 animals dead at birth showed lymphoid atrophy. The degree of tolerance was greater and runt disease more frequent and severe among those treated at the twentieth day of fœtal life than with those treated at the twenty-second day. The animals which developed a high degree of tolerance without showing runt disease clinically gained weight more slowly than their less tolerant litter mates and showed some lymphoid damage.

Some of the rabbits in which runt disease was induced developed an immune hæmolytic anæmia. Investigations into this phenomenon and the histological changes found in association with the runting syndrome will be reported in detail elsewhere.

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[1] Billingham, R. E., Brent, L., and Medawar, P. B., *Nature*, **172**, 603 (1953).
[2] Hašek, M., *Csl. Biol.*, **2**, 25 (1953).
[3] Woodruff, M. F. A., and Simpson, L. O., *Brit. J. Exp. Path.*, **36**, 494 (1955).
[4] Billingham, R. E., and Brent, L., *Transpl. Bull.*, **4**, 67 (1957).
[5] Billingham, R. E., Brent, L., and Medawar, P. B., *Phil. Trans.*, B, **239**, 357 (1956).
[6] Stark, R. B., Brownlee, H., and Grunwald, R. P., *Ann. N.Y. Acad. Sci.*, **73**, 772 (1958).
[7] Medawar, P. B., *J. Anat.*, **78**, 176 (1944).

VIROLOGY

Electron-dense Crystallites in Nuclei of Human Amnion Cells infected with Measles Virus

Measles virus antigen has been specifically identified in the nuclei of infected cells by means of the fluorescent antibody technique; and the development and spread of virus antigen in amnion and other human cells have been correlated with observations made in a concurrent study of fine structure[1]. During a continuation of the latter investigation we have found that crystallites made up of ordered arrays of electron-dense particles occur within nuclei of infected human amnion cells, and have also observed extracellular particles that can be interpreted as mature virus.

Twenty-one days after the cells were infected with the Edmonston strain of measles virus, they were fixed in osmium tetroxide, and ultra-thin sections were prepared for electron microscopy. Sections were mounted on 200-mesh grids and floated for 15 min. on a saturated solution of uranyl acetate to enhance contrast.

Many of the cells contained intracytoplasmic inclusion bodies that have been described in detail[1]. In addition, the nuclei of a few cells contained

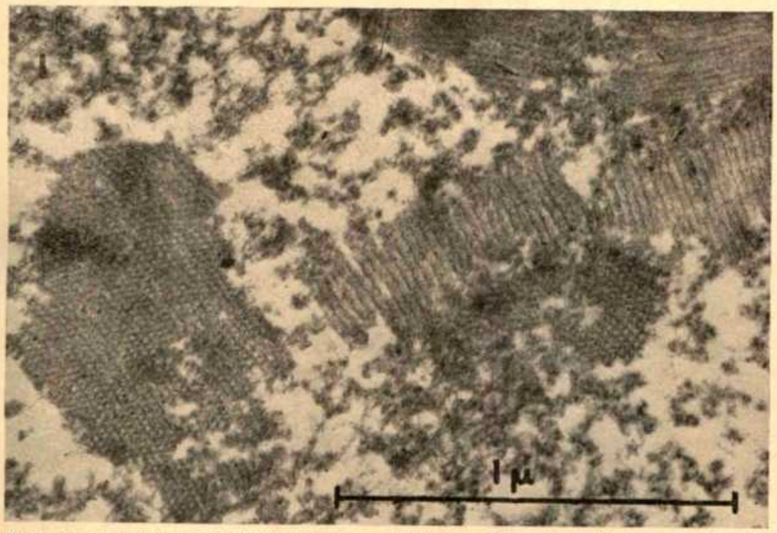

Fig. 1. Several orientations of intranuclear crystallites in a thin section of human amnion cell 21 days after infection with measles virus

crystalline arrays (Fig. 1). Crystalline arrays were not seen in uninfected amnion cells. It is reasonable to suppose that the arrays are associated in some way with virus multiplication. Three crystallographic directions can be identified in the crystallite at the left of Fig. 1, suggesting that a (111) plane is being visualized. The observed spacings along the three directions, in conjunction with the angles enclosed (70°, 70°, 40°), can be interpreted in terms of a unit cell with sides 120 Å. × 120 Å. × 211 Å. Other orientations are displayed by the crystallites at the right of Fig. 1, the spacings of which also are in agreement with the above interpretation.

It is unlikely that the crystallites consist of mature measles virus particles. Many of the cells containing crystalline arrays, or characteristic cytoplasmic inclusion bodies[1], or both, show particles on or outside the cell wall that might be interpreted as mature virus because of their morphology and size (Fig. 2). They exhibit double membranes, separated by 50 Å. ; display an internal granular structure ; and have an approximate diameter of 1200 Å. Benyesh *et al.*[2] have reported that the diameter of mature measles virus is approximately 1400 Å. Many of the bodies are 5- or 6-sided in outline, suggesting a polyhedral shape (Figs. 2 and 3).

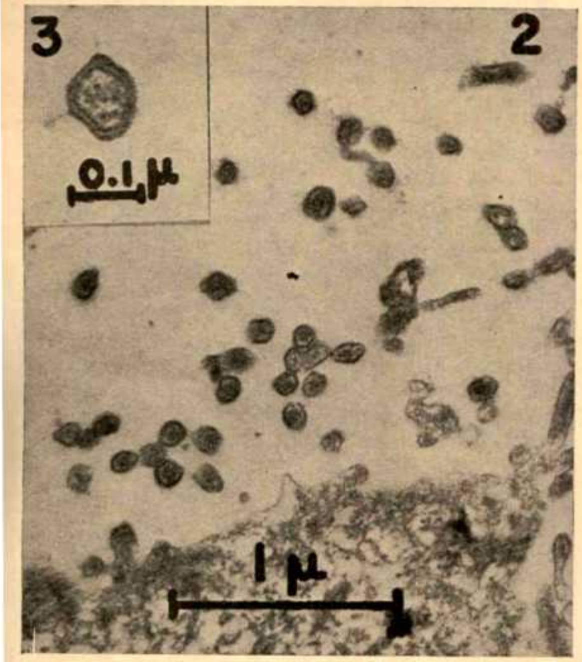

Fig. 2. Virus-like particles outside human amnion cell, showing internal structure and external membrane

Fig. 3. Higher magnification of a particle showing double membrane and suggesting polyhedral shape

Intranuclear crystallites have been observed in electron micrographs of animal cells infected with insect viruses and several mammalian viruses, including adenovirus[3] and herpes virus[4]. In some of the former and both the latter infections, the crystals appeared to be made up of the virus particles themselves. In addition, one type of crystal found in a type 5 adenovirus infection of HeLa cells was shown to be protein[4]. Morgan *et al.*[4] have discussed conditions that may favour the development of intracellular crystals in cells infected with virus, and speculate that the time-rate of virus assembly per unit volume may be the chief factor determining whether an ordered array will form.

Two mature animal viruses having polyhedral shapes have been demonstrated in electron micrographs. *Tipula* iridescent virus is 6-sided in cross-section and is icosahedral[5]. Adenovirus type 5 is also an icosahedron[6]. Three-dimensional studies are needed to determine which polyhedral shape might be assumed by mature measles virus.

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[1] Rapp, F., Gordon, I., and Baker, R. F. (submitted for publication). Baker, R. F., Rapp, F., and Gordon, I. (submitted for publication).

[2] Benyesh, W., Pollard, E. C., Opton, E. M., Black, F. L., Bellamy, W. D., and Melnick, J. L., *Virology*, **5**, 256 (1958).

[3] Harford, C. G., Hamlin, A., and Parker, E., *Trans. Assoc. Amer. Phys.*, **68**, 82 (1955). Morgan, C., Howe, C., Rose, H. M., and Moore, D. H., *J. Biophys. and Biochem. Cytol.*, **2**, 351 (1956).

[4] Morgan, C., Godman, G. C., Howe, C., and Rose, H. M., *Trans. Assoc. Amer. Phys.*, **81**, 281 (1958).

[5] Williams, R. C., and Smith, K. M., *Biochim. Biophys. Acta*, **28**, 464 (1958).

[6] Valentine, R. C., and Hopper, P. K., *Nature*, **180**, 928 (1958). Horne, R. W., Brenner, S., Waterson, A. P., and Wildy, P., *J. Molec. Biol.*, **1**, 84 (1959).

SOIL SCIENCE

Removal of Fixed Ammonium from Clay Minerals by Cation Exchange Resins

Ammonium in clay minerals that is not replaceable by K^+ has been defined as fixed NH_4^+. This fixed NH_4^+ is replaceable to some extent by Na^+ and H^+, but the amount that can be replaced depends upon the extraction method used. Recent work[1,2] has shown that small amounts of K^+ or NH_4^+ in the extracting solution block the replacement of fixed NH_4^+. Thus, the efficiency of an equilibrium extraction of fixed NH_4^+ is decreased by the accumulation of displaced NH_4^+ in the extracting solution[2]. If a clay mineral containing fixed NH_4^+ were equilibrated with a cation exchange resin instead of a soluble salt, there would be less soluble K^+ or NH_4^+ in the system. A comparison of these extraction methods was made, therefore, to determine if there was also less blocking and thus more fixed NH_4^+ removed in a resin extraction.

Samples of Montana vermiculite (0·2 gm.), Wyoming bentonite (0·3 gm.) and Goose Lake Grundite (illite, 0·5 gm.) that had been NH_4^+-saturated and dried at 110°, 350° and 110° C., respectively, were equilibrated with Na^+-, K^+- and H^+-saturated samples of 'Amberlite *IR*-120' in 20 ml. of distilled water. In each case $>$ 32 mesh resin material with an exchange capacity of 4·32 m.equiv. per gm. and a clay mineral : resin ratio of 1 : 20 (oven dry-weight basis) were used. Samples of the dried, NH_4^+-saturated clay minerals (0·2 gm. vermiculite, 0·3 gm. bentonite and 0·5 gm. illite) were also equilibrated with 20 ml. of 1 *N* sodium chloride and with 20 ml. of 1 *N* potassium chloride. The various systems were equilibrated at room temperature by shaking for periods of 2–8 days. The mixtures were then filtered, the residues were leached with 350 ml. of 1 *N* potassium chloride, and the NH_4^+ in the leachates was determined by distillation with potassium hydroxide. Before ammonium saturation, the illite was treated with a solution of sodium chloride and $NaB(C_6H_5)_4$ in order to remove K^+ and increase the NH_4^+ fixing capacity[3]. Illite samples that were not NH_4^+ saturated were included in each experiment, and the results obtained with the NH_4^+-saturated illite were corrected for nitrogen arising from that originally present in this material.

Table 1 gives the total NH_4^+ contents of the minerals (Kjeldahl analysis), their exchangeable NH_4^+ contents as determined by distilling 1-gm. samples with 400 ml. of 1 *N* potassium hydroxide, and the amounts of NH_4^+ removed by the resin and soluble salt extractions. In these extractions enough replacing cation was used to remove at least 80·6, 49·5 and 25·1 m.equiv. of NH_4^+ per 100 gm. from the vermiculite, bentonite and illite samples, respectively, if the distribution coefficient for the exchange of cations was 1.

Table 1. Amounts of NH_4^+ removed from NH_4^+-saturated Clay Minerals by Various Treatments (m.equiv. per 100 gm.)*

Treatment	Ammonium vermiculite	Ammonium bentonite	Ammonium illite
Kjeldahl digestion	81·4	50·4	25·3
Hydrogen resin	80·5	36·7	23·9
Sodium resin	80·5	36·3	23·2
Sodium chloride	66·5	33·0	22·4
Potassium resin	10·7	9·0	22·7
Potassium chloride	8·1	5·7	22·1
Potassium hydroxide distillation	2·8	24·6	22·6

* Mean of two or more determinations. The illite values are corrected for the NH_4^+ removed from similarly treated illite samples that were not NH_4^+-saturated.

Comparison of the results obtained by the resin and soluble salt extractions using the same replacing cation shows that although the resin method of extraction was the more effective with all three minerals, it was not markedly so except in the Na^+ treatment of vermiculite. When the NH_4^+ vermiculite was equilibrated with Na^+ (or H^+) resin essentially all of the NH_4^+ was removed, whereas only 76 per cent of the fixed NH_4^+ (that is, NH_4^+ not removed by distillation with potassium hydroxide) was removed by the sodium chloride treatment. It seems safe to assume that with this mineral the efficiency of the resin technique was due to a decrease in the blocking effect of the replaced NH_4^+, which limited extraction by the sodium chloride technique, since other experiments (to be reported later) indicate that it was not due to a selective adsorption of NH_4^+ by the resin or to a contact effect of the resin.

The influence of the resin on the removal of fixed NH_4^+ from illite and bentonite was very small, and essentially the same, whether Na^+ or K^+ was used. This suggests that with these minerals the effect of the resin was not due to a decrease in the blocking effect of displaced NH_4^+. More likely it was due to grinding of clay particles by the resin during the prolonged shaking period. This would also explain the observation that potassium resin extracted more NH_4^+ from vermiculite than did potassium chloride. In the vermiculite–resin extractions a distinct decrease in the size of the clay particles was observed.

The results in Table 1 show that the different treatments with K^+ (potassium hydroxide, potassium chloride, potassium resin) did not remove the same amount of NH_4^+ from the clay minerals. To define fixed ammonium, it is necessary, therefore, to specify the method of determination even when K^+ is used as the replacing cation. In the present work NH_4^+ that was not liberated as ammonia by distillation with potassium hydroxide was arbitrarily considered as being fixed.

Previous experiments with vermiculite have shown that the blocking effect of NH_4^+ or K^+ can be reduced by increasing the acidity of the extracting solution[2]. In the case of bentonite and illite, however, hydrogen resin was no more effective than sodium resin (Table 1). Also, instead of the increase observed with vermiculite, there was a decrease in the amount of NH_4^+ removed when the bentonite and illite samples were extracted with potassium chloride instead of potassium hydroxide. These results suggest that factors not encountered with vermiculite are involved in the fixation and release of NH_4^+ in bentonite and illite.

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[1] Hanway, J. J., *et al.*, *Soil Sci. Soc. Amer. Proc.*, **21**, 29 (1957).

[2] Scott, A. D., *et al.*, *Soil Sci. Soc. Amer. Proc.*, **22**, 388 (1958).

[3] Hanway, J. J., *Iowa State Coll. J. Sci.*, **30**, 374 (1956).

FORTHCOMING EVENTS

*(Meetings marked with an asterisk * are open to the public)*

Monday, March 14

University of London (at the Postgraduate Medical School of London, Ducane Road, London, W.12), at 4 p.m.—Prof. E. Zaimis: "Neurochemical Pharmacology".*

Institution of Electrical Engineers, London Graduate and Student Section (at Savoy Place, London, W.C.2), at 6 p.m.—Informal Meeting on "Should Engineers be encouraged to take up Administrative Positions early in their Careers?".

Tuesday, March 15

University of London (at Queen Mary College, Mile End Road, London, E.1), at 1.30 p.m.—Prof. C. D. Darlington, F.R.S.: "Heredity and Human Society".*

British Computer Society, London Branch (at the Northampton College of Advanced Technology, St. John Street, London, E.C.1), at 2.30 p.m.—Mr. N. Doveton: "Axis Transformation using Analogue Equipment".

University of London (at University College Hospital Medical School, University Street, London, W.C.1), at 4.30 p.m.—Prof. M. Maizels: "Salt and Water Balance in Cells".*

Royal Society of Arts, Commonwealth Section (at John Adam Street, Adelphi, London, W.C.2), at 5.15 p.m.—Sir Alan Burns: "Recent Developments in Fiji".

Institution of Electrical Engineers, Measurement and Control Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Mr. I. C. Hutcheon and Mr. D. Summers: "Fast-Response Transistor Chopper-Type Amplifier with Low Carrier Frequency".

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Prof. H. B. Squire, F.R.S.: "Some Problems of Separated and Vortex Flow".

Tuesday, March 15—Thursday, March 17

Oil and Colour Chemists' Association (in the Royal Horticultural Society's New Hall, Westminster, London, S.W.1)—Twelfth Technical Exhibition

Wednesday, March 16

Royal Society of Arts (at John Adam Street, London, W.C.2), at 2.30 p.m.—Dr. D. Roy Davies: "Induced Mutation in Crop Plants—Prospects and Prospectives" (Fernhurst Lecture).

Geological Society of London (at Burlington House, Piccadilly, London, W.1), at 5 p.m.—Dr. D. V. Ager: "The Epifauna of a Devonian Spiriferid"; Dr. G. Kelling: "The Stratigraphy and Structure of the Ordovician Rocks of the Rhinns of Galloway".

Royal Meteorological Society (at 49 Cromwell Road, London, S.W.7), at 5 p.m.—Mr. D. W. Martin and Mr. A. W. Brewer: "A Synoptic Study of Day-to-Day Changes of Ozone over the British Isles"; Mr. E. B. Kraus: "Synoptic and Dynamic Aspects of Climatic Change".

Royal Statistical Society (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.15 p.m.—Mr. P. A. Stone: "Economics of Building Designs".

Institution of Electrical Engineers, Supply Section (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. D. H. Grindell: "An Electrostatic Dust Monitor".

Society of Chemical Industry, Corrosion Group (at 14 Belgrave Square, London, S.W.1), at 6 p.m.—Miss D. M. Brasher: "The Mechanism of Inhibition in Neutral Aqueous Solutions".

Society of Chemical Industry, London Section (joint meeting with the London Section of the Royal Institute of Chemistry, at University College, Gower Street, London, W.C.1), at 6.30 p.m.—Prof. F. D. Richardson: "High Temperature Reactions Involving Condensed Phases".

Society for Psychical Research (at Kensington Town Hall, Kensington High Street, London, W.8), at 7 p.m.—Mr. H. Norman Hunt will reproduce recordings from Spiritualist Mediums.

Society of Instrument Technology (at Manson House, 26 Portland Place, London, W.1), at 7 p.m.—Mr. A. H. Doveton and Mr. K. C. W. Pedder: "The Simulation of a Large Chemical Plant on an Electronic Analogue Computer".

Pharmaceutical Society of Great Britain (at 17 Bloomsbury Square, London, W.C.1), at 7.30 p.m.—Dr. A. L. Glenn: "Hydrogen Ions and the Pharmacist".

Thursday, March 17

Royal Society (at Burlington House, Piccadilly, London, W.1), at 4.30 p.m.—Mr. A. R. Peacocke and Mr. B. N. Preston: "The Action of γ-Rays on Sodium Deoxyribonucleate in Solution. II Degradation. III: Denaturation"; Mr. J. Seed: "Inhibition of Nucleic Acid Synthesis Caused by X-irradiation of the Nucleolus".

Linnean Society of London (at Burlington House, Piccadilly, London, W.1), at 5 p.m.—Dr. V. H. Heywood: "Progress in the *Flora Europaea* Project"; Mr. Eric M. Marsden-Jones: "The Life Cycle of *Adleria kollari* Hartig (Cynipidae)".

London Mathematical Society (at the Royal Astronomical Society, Burlington House, Piccadilly, London, W.1), at 5 p.m.—Dr. P. J. Higgins: "Two Methods in Combinational Analysis".

Institute of Physics, London and Home Counties Branch (at 47 Belgrave Square, London, S.W.1), at 6 p.m.—Prof. F. P. Bowden, C.B.E., F.R.S.: "The Friction and Deformation of Solids at Very High Speed".

Royal Aeronautical Society (at the Institution of Mechanical Engineers, 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Mr. M. B. Morgan: "Supersonic Aircraft—Promise and Problems.".

Society of Chemical Industry, Road and Building Materials Group (at 14 Belgrave Square, London, S.W.1), at 6 p.m.—Mr. P. H. Robin: "Floors Old and New".

Royal Society of Tropical Medicine and Hygiene (at the Royal Army Medical College, Millbank, London, S.W.1), at 7.30 p.m.—Laboratory Meeting. Chairman Sir William MacArthur, K.C.B., D.S.O., M.D., F.R.C.P.

British Institute of Radiology, incorporated with the Rontgen Society (at 32 Welbeck Street, London, W.1), at 8 p.m.—Meeting on "The Reduction of Radiation Hazards in the Therapeutic Use of Small Multiple Radioactive Sources".

Friday, March 18

British Psychological Society, Occupational Psychology Section (in the Department of Psychology, Birkbeck College, Malet Street, London, W.C.1), at 1 p.m.—Dr. Marie Jahoda: "Some Remarks on the Study of Technological Education".

University of London (at the Postgraduate Medical School of London, Ducane Road, London, W.12), at 4 p.m.—Dr. J. A. Fraser Roberts: "Medical Genetics".*

Royal Society of Medicine, Epidemiology and Preventive Medicine Section (at 1 Wimpole Street, London, W.1), at 5.30 p.m.—Dr. W. R. S. Doll and Dr. W. M. Court Brown: "Studies on the Possible Effects of Low Doses of Radiation".

Institution of Electrical Engineers, London Graduate and Student Section (joint meeting with the Graduate and Student Section of the Institutions of Civil and Mechanical Engineers, at the Institution of Mechanical Engineers, Birdcage Walk, London, S.W.1), at 6.30 p.m.—Address by Sir George Edwards, C.B.E.

Society for Analytical Chemistry (joint meeting with the Fine Chemical Group of the Society of Chemical Industry, at the School of Pharmacy, Brunswick Square, London, W.C.1), at 6.30 p.m.—Meeting on "Techniques of Automatic Analysis". Mr. R. M. Pearson: "Automatic Analysis in the Chemical Industry"; Dr. I. D. P. Wootton: "Automation in Clinical Biochemistry".

Antiquarian Horological Society (at the Science Museum, South Kensington, London, S.W.7), at 7 p.m.—Discussion on the work of the Ellicotts.

British Institute of Radiology, incorporated with the Rontgen Society (at 32 Welbeck Street, London, W.1), at 8 p.m.—Meeting on "Tritium Counting".

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Prof. R. V. Jones: "Infra-red Radiation".

Saturday, March 19

Nutrition Society (at the Medical College of St. Bartholomew's Hospital, Charterhouse Square, London, E.C.1), at 9.45 a.m.—Symposium on "Nutrition and the Elderly".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Senior Lecturer in Hydro- and Aero-dynamics; and a Senior Lecturer in Physics—The Registrar, College of Advanced Technology, Gosta Green, Birmingham 4 (March 19).

Assistant Lecturer in the Department of Mathematics—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1 (March 22).

Lecturer in Applied Mathematics—Joint Clerk to the University Court, Queen's College (University of St. Andrews), Dundee (March 26).

Lecturer (with medical or scientific qualifications) in Physiology—Joint Clerk to the University Court, Queen's College (University of St. Andrews), Dundee (March 26)

Research Fellows (Ph.D. standard, with a special aptitude for research in high or low energy nuclear physics) in Experimental or Theoretical Nuclear Physics—The Registrar, The University, Liverpool (March 26).

Leverhulme Research Fellow (graduate in medicine or science)—The Secretary, St. Mary's Hospital Medical School, Paddington, London, W.2 (March 28).

Research Fellow (with research experience in biochemistry or chemistry, and an interest in the physico-chemical aspects of biochemistry) in the Department of Physical Biochemistry, John Curtin School of Medical Research, Australian National University—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (March 30).

Assistant Experimental Officer (preferably woman aged at least 22, with B.Sc. pass degree, with bacteriology as principal or subsidiary subject, or equivalent) at Torry Research Station, D.S.I.R., Aberdeen, to work on the maintenance of type cultures for the National Collection of Industrial Bacteria—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. G.68/0A (March 31).

Assistant Lecturer in Philosophy, tenable in either the Department of Philosophy, or in the Department of Political and Moral Philosophy—The Registrar, University College of North Staffordshire, Keele, Staffs (March 31).

Chair of Physics at University College, Ibadan, Nigeria—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 31).

Demonstrators (2) (graduates) in Agriculture, for duties on the University farms—The Registrar, The University, Leeds 2 (March 31).

Foulerton Gift Research Fellow (British nationality) for original research in Medicine—The Assistant Secretary, The Royal Society, Burlington House, Piccadilly, London, W.1 (March 31).

Junior Lecturer in Botany—The Registrar, Trinity College, Dublin (March 31).

Lecturer or Assistant Lecturer in Mechanical Engineering—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1 (March 31).

Lecturer or Demonstrator in Agricultural Biochemistry—The Registrar, King's College (University of Durham), Newcastle upon Tyne 1 (March 31).

Research Fellow in the Biophysics Laboratory, Department of Physics, for work largely concerned with the electric properties of plant membranes and tissues and the relation of these to plant development—The Registrar, University of Tasmania, Box 252O, G.P.O., Hobart, Tasmania (March 31).

Senior Lecturer or Lecturer in Mathematics at the University of Melbourne, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (March 31).

White's Professor of Moral Philosophy—The Registrar, University Registry, Oxford (March 31).

Research Worker in Psychopharmacology for work concerned with the effects of psychotropic drugs on psychological and physiological functions, and will involve studies on normal subjects and psychiatric patients—Dr. H. O. Schild, Department of Pharmacology, University College, Gower Street, London, W.C.1 (April 2).

Chair of Biochemistry—The Registrar, The University, Leicester (April 8).

Lecturer in Social Anthropology; and a Lecturer in Agricultural Chemistry—The Secretary of University Court, The University, Glasgow (April 11).

Lecturer in Chemistry in the Department of Chemistry and Chemical Engineering, University of Natal, Durban—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa and London, April 15).

Lecturer or Assistant Lecturer in Zoology at the University of Malaya (Singapore Division)—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (April 15).

Lecturer/Senior Lecturer (qualified in grassland management) in Animal Husbandry at the University of Sydney Animal Husbandry Farms, Camden, New South Wales—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, April 15).

Lecturer (with research interests in either branch) in Inorganic and/or Physical Chemistry; a Lecturer in Mathematics; and a Lecturer in Moral Philosophy in St. Salvator's College—Joint Clerk to the University Court, College Gate, St. Andrews (April 16).

Assistant Lecturer (with special qualifications in the genetics of micro-organisms) in the Sub-Department of Genetics—The Registrar, The University, Liverpool, quoting Ref. CV/T (April 19).

Research Fellow in Pure Science; a Research Assistant (preferably with a special interest in educational psychology and/or general experimental psychology) in Psychology; and a Graduate Assistant in Zoology, with special responsibility for the Departmental Museum—The Secretary, The University, 38 North Bailey, Durham (April 23).

Senior Lecturer (with a postgraduate degree in either geography or geology from a British university or the equivalent, and teaching experience at university level) in Geography and Geology at the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, April 30).

English Electric Research Fellow in Aeronautics, for postgraduate research into the problems of vibration or stability and control of aircraft—The Warden, The College of Aeronautics, Cranfield, Bletchley, Bucks (May 1).

Professor of Engineering Science—The Registrar, University Registry, Oxford (May 5).

Biochemist (B.Sc. or preferably Ph.D., with experience in bacteriology)—The Secretary, Wright Fleming Institute, St. Mary's Hospital Medical School, Paddington, London, W.2.

Biochemist, Senior grade, to take charge at East Riding Group Laboratory—The Secretary, Westwood Hospital, Beverley, East Yorkshire.

Experimental Officers/Assistant Experimental Officers (with G.C.E. "A" level in chemistry and another science or mathematics subject or equivalent, but if over 22, a pass degree, H.N.C. in chemistry or equivalent is generally expected) at the National Chemical Laboratory, D.S.I.R., Teddington, Middx., for work on general inorganic chemistry, microanalysis, electro-chemistry, chemical thermodynamics and corrosion—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1 quoting Ref. F.123/0A.

Hospital Physicist to the Radiotherapy Department—The House Governor, St. Mary's Hospital, Paddington, London, W.2.

Laboratory Technician in the Agricultural Botany Section of the Department of Agricultural Sciences—The Secretary, University of Nottingham, School of Agriculture, Sutton Bonington, Loughborough.

Lecturer or Assistant Lecturer (man or woman graduate, preferably with teaching experience) in Physics in the Training Department—The Warden, Goldsmiths' College (University of London), New Cross, London, S.E.14.

Mathematicians (with a university degree and experience in working with digital computers) with the European Organization for Nuclear Research—CERN, Geneva, for the analysis and programming of various computing problems, in particular problems of numerical analysis on a Ferranti Mercury and IBM 704—The Ministry of Labour (E.9), 26–28 King Street, London, S.W.1.

Physical Chemists (with a good honours degree and preferably some postgraduate research experience), to carry out fundamental work on the reaction between metals and water at high temperatures—The Director, Arthur D. Little Research Institute, Inveresk, Midlothian, Scotland.

Research Assistant (graduate, preferably with experience in microanalytical techniques and chromatography), for work on biosynthesis and identification of steroid hormones in the adrenal gland—Mr. P. J. Ayres, Barnato Joel Laboratories, The Middlesex Hospital, Mortimer Street, London, W.1.

Research Assistant (honours B.Sc. with experience in blood grouping, biochemistry, bacteriology or mammalian genetics), for work on transplantation immunity, including mouse blood group genetics, C.F., skin transplantation—Dr. H. B. Amos, Roswell Park Memorial Institute, Buffalo 3, New York, U.S.A.

Scientific Officer (graduate, preferably in physics, and capable of maintaining and developing the heart lung machine and electronic and other research apparatus) to assist in developments related to cardiac surgery—The Assistant Administrator, Radcliffe Infirmary, Oxford.

Senior Scientific and Scientific Officers (with a first- or second-class honours degree or equivalent in science, engineering or mathematics), for a very wide range of research and development work, including solid state physics, radio astronomy, advanced computing techniques, and electronics and radar—The Director, Royal Radar Establishment, Malvern, Worcs.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

Department of Scientific and Industrial Research: Road Research Laboratory. Road Note No. 24: Roadstone Test Data presented in Tabular Form. Pp. 8. (London: H.M. Stationery Office, 1959.) 9*d*. net. [3012

Laboratory Animals Centre. Collected Papers, Vol. 8 (1959): Report of a Symposium on Quality in Laboratory Animals, held at the Royal Veterinary College, London, on 16th April, 1959. Pp. 68. (Carshalton: Laboratory Animals Centre, M.R.S. Laboratories, 1959.) 10*s*. [3012

Department of Scientific and Industrial Research. Food Investigation 1958: Reports of the Superintendents of the Low Temperature Research Station and the Ditton Laboratory. Pp. vi+42. (London: H.M. Stationery Office, 1959.) 3*s*. net. [3012

Society for the Promotion of Nature Reserves. Handbook 1959: Forty Second Annual Report, year ending 31st March, 1959. Pp. 56+4 plates. (London: The Society for the Promotion of Nature Reserves, c/o British Museum (Natural History), 1959.) 10*s*. [3012

Other Countries

Arbeitsgemeinschaft für Forschung des Landes Nordrhein-Westfalen Heft 78: Experimentelle Studien über die Distanzeffekte in Bestrahlten Vielzelligen Organischen. Von Fréderic Ludwig. Die Anwendung Radioaktiver Isotope in der Chemischen Forschung. Von A. H. W. Aten, Jr. Pp. 88. (Köln und Opladen: Westdeutscher Verlag, 1959.) 9.20 D.M. [3012

The Story of Chemical Industry in Basle. Pp. 234. (Published by CIBA Limited on the Occasion of its 75th Anniversary.) (Basle: CIBA Limited, 1959.) [3012

Chicago Natural History Museum. Fieldiana: Zoology. Vol. 39, No. 31 (May 22, 1959): Inland Mollusks from Venezuela, Southern Brazil, and Peru. By Fritz Haas. Pp. 363–371. (Chicago: Chicago Natural History Museum, 1959.) 30 cents. [3012

Profiles of the Time of James Monroe, 1758–1831: an Exhibition Commemorating the 200th Anniversary of the Birth of the Fifth President of the United States, October 26–November 23, 1958. Pp. 32. (Publication 4348.) (Washington, D.C.: Smithsonian Institution, 1959.) [3012

Smithsonian Miscellaneous Collections. Vol. 139, No. 5: Genera of Tertiary and Recent Rhynchonelloid Brachiopods. By G. Arthur Cooper. Pp. iii+90+22 plates. (Publication 4382.) (Washington, D.C.: Smithsonian Institution, 1959.) [3012

Mitteilungen der Prähistorischen Kommission der Österreichischen Akademie der Wissenschaften. VIII und IX Band: Willendorf in der Wachau: Monographie der Paläolith-Fundstellen I–VII. 1 Teil: Text. Pp. v+219. 2 Teil: Inventar. Pp. 79. 3 Teil: Abbildungen. Abb. 124. (Wien: Österreichische Akademie der Wissenschaften, 1959.) [3012

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ACADEMIC FREEDOM AND UNIVERSITY STANDARDS

IN contrast to the protests, so far unsuccessful, against the infringement of university autonomy in South Africa by the Union Government's Extension of University Education Act, which is universally recognized throughout the academic world as striking against the fundamental conception of a university, a recent *Bulletin of the Committee on Science and Freedom* (No. 14; February 1960) records a successful outcome to a storm of protest from academic and public opinion in Queensland. This was against a Government measure which attempted to establish an officially sponsored appeal board through which the Government could control university staff appointments. This outcome is perhaps the more encouraging in that it demonstrated the existence of an unexpected number of allies in a community where generally graduates—and also scientists, as the Murray Committee's report on Australian universities has shown—are still a small minority and one not held in high enough repute. It should be noted, however, that freedom of platform and Press is far more of a reality in Australia than in South Africa, and this, as Prof. T. K. Ewer shows in the article referred to above, is an important factor when a vital liberty is threatened.

Prof. Ewer describes how the threat arose when the Government of Queensland introduced in 1957 the University of Queensland Acts Amendment Act, 1957. Since the eight universities in Australia, except those of Sydney and Melbourne, are almost wholly dependent on Government funds, the legislation under which they operate seeks to assure strong Government representation on the governing bodies. This Act incorporated a number of amendments which were acceptable to the Staff Association and to the Senate, although a request of the former for three representatives on the Senate was unacceptable to that body, which nevertheless supported the principle of direct representation of staff. This, however, was limited to one member, and so far, in spite of the large expansion of university staff in the past decade and strong representations by the University Staff Association to the Minister of Education, this number has not been increased to the suggested three. In fact, the lack of sympathy shown by Government representatives on the Senate to such matters as an acceptable superannuation scheme, staff housing and the maintenance of parity in salary scale with the Universities of Sydney and Melbourne emphasize both the importance of an informed and alert public opinion and of the comments of the Murray Committee. The expenditure by the Australian Government in the next twelve years of £A20 million recommended by the Murray Committee will not by itself suffice to remove the shortage of scientists and technologists in Australia in academic or other employment, unless their work is appreciated by the community and paid for in accordance with its value.

The real threat described by Prof. Ewer, however, arises from Sections 25 and 31 in the Act, which stated the principle of allowing appeals, to be made to a specially constituted appeal board, against academic and non-academic appointments made by the University and also against disciplinary action. This provision was not to apply to the offices of vice-chancellor or registrar, but the composition of the appeal board raised the main criticism and opposition within and without the University. This was to consist of three members: one appointed by the Government, who was to be chairman; one appointed by the Senate; and one appointed by the Staff Association. These provisions caused the most vigorous protest throughout Australia; it is not quite clear how the blunder was made, but the processes of democracy saved the day.

Without going into the full details, it may be noted that a public meeting of some 2,400 people in the City Hall, Brisbane, passed with acclamation a resolution stating that the provision of an appeal board is "a direct threat to academic freedom and to academic standards of the university; departs from the tradition and procedures of universities within the British Commonwealth; is a great disservice to the interests of the people of Queensland". Pointed reference was made at the meeting to the threat which political interference with academic appointments offered to recruitment, particularly for professorships and senior posts, where reliance is still largely on overseas applicants. The defeat of the Government in a general election, however, and return of a new Government to power was followed by repeal of the appeal board provisions on December 17, 1958, and the new University of Queensland Acts Amendment Act, omitting the appeal board provisions, was proclaimed on August 11, 1958.

This unexpected attack on the University's autonomy has been followed by much discussion among the University staff, and the Professorial Board reconsidered the methods hitherto used for making appointments at all levels. Proposals were framed to ensure that every applicant for a new post on the University staff would be interviewed by the selection committee; the existing system of committees and referees is much the same as that employed in other British universities. The vital lesson of the episode, however, seems to be the importance of public understanding and the existence of effective democratic institutions.

It is the absence of these in South Africa that has been responsible for the passing of the unfortunate Extension of University Education Act and for its operation. Already eight members of the staff of the University College of Fort Hare have been dismissed by the South African Government under the Act, without reason being given; and it seems clear that opposition to government policy will now be regarded as grounds for dismissal and that the members of staff thus dismissed, who include the registrar, five professors, the librarian and a lecturer, are the first victims of the political control of university institutions for non-white peoples. Speaking at the meeting of the Tunis study group of the Committee on Science and Freedom on "Freedom and Responsibility: the Role of the Scholar in Society" in April 1959, Dr. E. G. Malherbe maintained that the idea of a university completely controlled by the State was repugnant to South African men who had learned to value the traditional British system of freedom and autonomy. While, however, the Act would affect adversely the recruitment of staff of good quality and thus tend to debase South Africa's academic currency, its most serious effect would be the exclusion of non-white students from existing universities. Unless the Government instituted a very elaborate system of bursaries, it was difficult to see how the millions of non-whites who are living outside the projected Bantustans, and are completely integrated into the economy of the white people, would have any chance to obtain a university education. Apart from considerations of autonomy and academic freedom, the issue would become economic. South Africa, for a civilized country, has a very low national income per head, and it is difficult to see how she could aspire to multiply university institutions merely in the interests of preserving the ethnic identity of peoples who have evolved no written language and whose culture is riddled with witchcraft and magic.

Dr. Malherbe was confident that, apart from this principle of exclusion in the new legislation, the academic freedom and autonomy of the existing universities of South Africa continue to stand as high as in any country in the world; but with these dismissals at Fort Hare before us it may well be doubted how long this will last. The free society anywhere depends for its strength upon the exercise of civic responsibility by its members. and though the academic community can play a major part by the manifestation of courage and strength of conviction in maintaining the vigorous growth of the ideal and reality of freedom, it cannot continue indefinitely to function as a community unless those qualities command sufficient respect among the public at large to ensure maintenance of the essential conditions of academic freedom of teaching, investigation and appointment. The conference at which Dr. Malherbe's paper was given provided encouraging evidence of the extent to which the idea of academic freedom has taken root, and at the present time it is particularly interesting to note this in the papers by Prof. B. A. Fletcher on the university and society in Central Africa, by Prof. E. Njoku on the relation between university and society in Nigeria, and by Dr. H. A. Oluwasanmi on the scholar and society in Nigeria.

Prof. Fletcher touched briefly on the way in which the University College of Rhodesia and Nyasaland, at Salisbury, is seeking to serve its society in respect of teaching, research and intellectual guidance, emphasizing that in industry, politics and religion it should leave leadership to others. It can both serve society and nourish itself by research and collaborative inquiry in the major fields of science, the social sciences and education, though these are not the only avenues. It was clear enough that Prof. Fletcher's vision of the service a university can render its society can only be realized in a university free from control by Government or any external agency, and free to determine for itself what it shall teach and whom it shall admit, freedoms which in general, as Dr. J. W. Cook observed in an introductory paper on university traditions with particular reference to the British Commonwealth, can only be safeguarded if the academic community has a large measure of control over appointments to its own body.

Prof. Njoku was optimistic that, given proper cultivation, academic freedom should root strongly in Nigeria, and Dr. Oluwasanmi, observing that the Western university tradition is still new in Nigeria, pointed out that, though Ibadan University College derives the bulk of its finances from the State, it has succeeded in maintaining its academic autonomy. It is essential, however, he said, that such university institutions in multi-racial societies should increasingly draw their inspiration from the environment in which they live and function if they are to accept the additional, if non-traditional, function of creating conditions from which a national, as distinct from tribal or racial, identity can be forged. This means that such institutions must also give more attention to those unexplored regions of knowledge that emphasize the common elements of the diverse cultures of the territories in which they function.

There was no tendency at this meeting to ignore the dangers which political and other conditions offer to the realization of these ideals: they were explicitly faced by both Prof. Njoku and Dr. Oluwasanmi, and equally by Dr. Cook, who put his finger on some of the obstacles which the surging tide of nationalism, for example, can raise. While it is easy to sympathize with the desire to foster the use of national languages, it can be an important factor in preventing the attainment of satisfactory standards in a university as well as hinder the free exchange of information and thought with the Western world, particularly in science, where a substantial proportion of original literature is published in English. A university has a duty to guard against any action which might weaken it in its pursuit of learning and the search for knowledge in the less advanced countries, no less than in the advanced and highly industrialized countries.

Dr. Cook said he is convinced that the policy adopted in 1945 in developing these new university institutions in the Commonwealth countries had been right. High academic standards have been established from the start, staffs of high calibre appointed, and significant and important contributions to new knowledge, often related to the needs and opportunities of the regions, have been made. There has also been a small but steadily increasing flow of graduates who will have a profound influence on the development of their nations.

All this will be of increasing importance as more territories move to independence and increasing efforts are made to solve the problems of government in multi-racial territories. More university institutions will probably be required, and Dr. Cook wisely suggests that these need not be conceived on precisely the same lines as the existing institutions. There is room for experiment, and perhaps especially in ways that have closer regard to the financial resources of newly independent countries. What matters most is that in giving careful attention to the traditional cultures of the countries and to their expanding needs, high academic standards should be maintained and the university traditions which have stood the test of time should be scrupulously observed. For that reason alone, universities in the rapidly developing countries should be the more careful not to become embroiled in political controversy, and to ensure that their legitimate studies of political, economic and social questions should be undertaken in no partisan spirit. Nor may it be too much to hope that in the measure in which these new universities meet the needs of the independent territories, and especially of the multi-racial communities, they will demonstrate the futility of the South African Government's present university policy, and perhaps even cause its reversal before South African academic traditions are destroyed and irreparable damage done to its universities.

ANTIBIOTICS FROM A NOVEL ANGLE

Antibiotics

Their Chemistry and Non-Medical Uses. Edited by Dr. Herbert S. Goldberg. Pp. x+608. (Princeton, N.J.: D. Van Nostrand Company, Inc., London: D. Van Nostrand Company, Ltd., 1959.) 112*s.* 6*d.*

In a foreword to this very welcome book, Dr. Paul R. Burkholder, the discoverer of chloromycetin, says: "For some time past, all of us have been certain about one aspect of antibiotic science and that is the great need for a new kind of book which would summarize the remarkable advances that have taken place recently" and the reviewer, who is in full agreement, would like to add "particularly in their non-medical uses". The book is the work of six American authors, all of whom are well known in antibiotic science. It consists of an introduction and six chapters, each of which is a complete monograph in itself. An unusual and very useful feature of the book is the large number of references—some two thousand in all—occupying a total of 261 pages and amounting to about 43 per cent of the book. The references given in the introduction (216 in number) and in the opening chapter on the chemistry of antibiotics (566) give the full title of the paper referred to in the text together with the usual details. In the remaining five chapters, however, very informative abstracts of the contents of the papers are also given.

The introduction deals with the history of antibiotics, their economic import, natural occurrence, effects and mode of action. The opening chapter (84 pages) admirably succeeds in summarizing the chemistry of the penicillins, streptomycin and related antibiotics, chloromycetin, the tetracycline group, the macrolides, novobiocin, polyene and polypeptide antibiotics.

The growth stimulation and general effects on the nutrition of domestic animals and birds is discussed in the following chapter (59 pages; 545 references). The magnitude and importance of this aspect of antibiotics may be judged from the fact that in 1954 more than 20 per cent (value 25 million dollars) of all antibiotics produced in the United States were used in animal feed supplements.

Botanists, and particularly plant pathologists and physiologists, will find the article on "The Influence of Antibiotics on Plants and Plant Disease Control" (78 pages; 376 references) a full and authoritative account of the subject. It is certainly surprising to learn that at least thirty-nine different antibiotics have been studied in the control of plant bacterial and fungal pathogens.

The texts of the remaining three chapters are relatively short. Chapter 5 (22 pages; 283 references, which unfortunately are not numbered) deals with antibiotics in food preservation, that is, in canned foods, fresh foods including meats, poultry, fish, fruit and vegetables, dairy products and yeast fermentations. Laboratory workers in microbiology will particularly welcome chapter 6 (18 pages; 96 references) on the uses of antibiotics in the isolation in pure culture and cultivation of micro-organisms from the complex mixtures in which they normally occur, for example, bacteria, fungi including plant pathogens, protozoa, viruses and rickettsiae. The book closes with an authoritative article (22 pages; 75 references) on the public health significance of non-medical uses of antibiotics, particularly in foods for human consumption. We may however take some comfort from the author's opinion that "one must conclude that the seriousness of the public health hazards of antibiotic residues in food has been overstated".

I believe that this book will completely satisfy the need expressed earlier for a new kind of book on antibiotic science. HAROLD RAISTRICK

BIOCHEMICAL PRACTICE

Biochemical Preparations

Vol. 6. Edited by Carl S. Vestling. Pp. ix+105. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1958.) 42*s.* net.

Methods of Biochemical Analysis

Vol. 7. Edited by Prof. David Glick. Pp. ix+353. (New York: Interscience Publishers, Inc.; London: Interscience Publishers, Ltd., 1959.) 72*s.*

THE latest volume in the valuable series of "Biochemical Preparations" contains detailed descriptions of carefully checked methods for the preparation of the following: crystalline animal cytochrome *c*, deoxyribonucleic acid, 2,3-diphosphoglyceric acid, L-α-glycerophosphorylcholine, 3-

hydroxyanthranilic acid, β-hydroxy-β-methylglutaric acid, insulin, lanosterol, leucine amino-peptidase, α-methylserine and *bis* (hydroxymethyl) glycine, crystalline horse oxyhæmoglobin, old yellow enzyme, crystalline papain and benzoyl-L-argininamide, crystalline muscle phosphorylases *a* and *b*, phosphoserine, rat liver ribonucleic acid, ribulose diphosphate, and DL-tryptophan-7a-C^{14}. A few minor criticisms are worthy of mention. The excellent practice of including structural formulæ was apparently overlooked in the description of the preparation of lanosterol. It is regrettable that anachronisms such as "Twenty-one and two-tenths grams of L-arginine hydrochloride . . ." (p. 64) and "Two and six-tenths kilograms of the washed and dried yeast . . ." (p. 55) are preferred to the more usual form. Grammatical errors, such as "The enzyme is extracted from dried brewer's bottom yeast, impurities removed by precipitation with basic lead acetate, and the enzyme further purified by fractionation . . ." (p. 54), and "The combined eluates are dialyzed and the adsorption procedure repeated . . ." (p. 57), were surprising.

The editorial board clearly makes strenuous efforts to replace older methods in earlier volumes by better methods, as illustrated by the inclusion of improved preparations of muscle phosphorylase and cytochrome *c*. There must be an interval before this is possible, however, and it is suggested that an appendix in each volume might give references to recent descriptions in the original literature of new methods of preparation of compounds, which have formed the subject-matter of earlier volumes of this series. One example will suffice; the preparation of leucine aminopeptidase has been simplified (Folk, Gladner, and Viswanatha, *Biochim. Biophys. Acta*, 36, 256; 1959) since this volume was published, and, although not checked by independent workers, reference to the method would be helpful in succeeding volumes. There is a valuable list of compounds of biochemical interest which have appeared in "Organic Syntheses" up to Vol. 38.

The volumes of "Methods of Biochemical Analysis" have rightly assumed a position of authority in biochemical literature. The latest volume deals with eight topics. Various modifications of the technique of immunoelectrophoresis are described in detail by P. Grabar, who is a pioneer in this field. This will please many workers, for much of the original literature appears in recondite journals. Two articles of considerable value to toxicologists describe the analysis of basic nitrogenous compounds (A. S. Curry) and the determination of ethyl alcohol in blood and tissues (F. Lundquist). The latter would provide the Minister of Transport with some interesting home-work. Workers in the lipid field are catered for by two chapters entitled "The Determination of Inositol, Ethanolamine, and Serine in Lipides" (J. M. McKibbin) and "The Assay of Lipoprotein Lipase *in vivo* and *in vitro* (E. D. Korn). L. B. Jaques and H. J. Bell have given a detailed description of the bewildering variety of methods for the extraction and determination of heparin. This article, however, contains the astonishing statement that "Barium due to its high molecular weight will dilute and hence reduce the values". The rather short account of the determination of creatinine and related guanidinium compounds (J. F. Van Pilsum) could have included a more critical discussion of the numerous colorimetric methods for arginine determination. Several recent methods (Satake and Luck, *Bull. Soc. Chim. Biol.*, 40, 1743; 1958: Cerriotti and Spandrio, *Biochem. J.*, 66, 603; 1957: Janus, *Nature*, 177, 529; 1956) are not mentioned. The recently developed technique for spectrophotometric study of translucent materials, which employs an opal glass in the light beam, is described by Shibata. Theoretical principles are considered, and suitable attachments for commercial spectrophotometers are described.

D. T. ELMORE

THE WAY TO THE STARS

Advances in Astronautical Sciences

Vol. 1: Proceedings of the Third Annual Meeting of the American Astronautical Society, December 1956. Pp. vi+177. Vol. 2: Proceedings of the Fourth Annual Meeting, December 1957. Pp. viii+456. Vol. 3: Proceedings of the Western Regional Meeting, August 1958. Pp. viii+547. Vol. 4: Proceedings of the Fifth Annual Meeting, November 1958. Pp. ii+460. (New York: Plenum Press, Inc.; London: Chapman and Hall, Ltd., 1957, 1958 and 1959.) 80*s*. net each volume.

ASTRONAUTICS, rejoicing in respectability and a far-flung dependence on numerous aspects of science and technology, finds itself admirably suited to congresses and conventions. The records of the proceedings tend to be impressive, expensive and distinctly daunting to the general reader, however great his specialist knowledge in one or two related fields.

The present volumes are no exception: they record the proceedings of four separate meetings of the American Astronautical Society held in December 1956 (Vol. 1), December 1957 (Vol. 2), August 1958 (Vol. 3) and November 1958 (Vol. 4). Each volume costs 8 dollars which, by some curious transformation process, becomes £4 in Great Britain. Taken together, they comprise 143 papers with some abstracts and supplementary lectures, covering 1,640 pages and weighing 8 lb. Vol. 1, which is less than half the size of any of the others but costs just as much, begins with a short paper by C. S. Draper on "Education in the Astronautical Sciences". He suggests a 4–6-year curriculum likely to overwhelm all but the most able and enthusiastic student. Many of the following papers are speculative rather than specific, and the volume ends with much discussion about the twin or clock paradox and relativity.

The remaining three volumes are all post-*Sputnik* 1 and reflect the enormous impetus that the launching of the world's first artificial satellite gave to American astronautics. An appreciable proportion of the latter volumes consists of sober theoretical analyses of the motions of space vehicles, communication and navigation problems, reports of full-scale and laboratory experiments and the development of ideas based on verified facts or current techniques. In all these matters the exposition is generally excellent, and we are—to use a somewhat inappropriate simile—on solid ground. Some of the remaining papers are futuristic and here the reviewers find themselves rather at a loss. In most fields of engineering, development is achieved by making an accurate estimate of the worthwhile magnitude of the next step forward, on technical and economic grounds. One can easily think of examples in aviation where the steps have been either too conservative or too bold. When reading about ideas in astronautics, the practical applications of which are often exceedingly complex but which advance very rapidly, it is difficult to judge just how far ahead the author may be thinking. It certainly seems that some of the brains devoted to contemplating future developments have taken their thoughts well beyond the next few feasible technological steps: in the absence of a yardstick, it becomes difficult to distinguish between far-ranging speculation or suggestion and something very close to science fiction. One can only marvel at the opulence of an industry capable of supporting gentlemen who devote their thoughts to "Assembly of a Multi-Manned Satellite" particularly on reading sentences such as: "Imagine then a space-suited worker chasing tools and materials on different orbits". Of course, such papers are intensely interesting, particularly when written in concise English, as is this particular example. Papers using turgid prose are fortunately in the minority.

One is therefore caused to question the object of all this activity, and is gratified to find that Dryden's contribution to the introduction of Vol. 4 is entitled "Space Exploration and Human Welfare". It is probably unfair to expect a concisely stated objective: the paper lists a few minor benefits to television and meteorology, says quite bluntly that we do not know the ultimate role of space vehicles in transportation and makes reference to the desire for increasing our knowledge of the universe for peaceful purposes. It seems quite possible that something is left unsaid.

The binding and paper of these volumes are excellent: unfortunately the reproduction of the contents is very variable in quality. The first three volumes consist of reproductions of typescript and handwritten equations. The diagrams reproduce well, but most of the photographs are very unsatisfactory. Volume 4 reverses this situation. The print and photographs are generally good but the diagrams, while clear, are reproduced on far too small a scale.

These volumes contain much valuable information and would be useful additions to a generously endowed library.

F. G. IRVING
J. L. STOLLERY

PLANT PHYSIOLOGY IN THE FIELD

Plants and Environment

A Textbook of Plant Autecology. By Prof. R. F. Daubenmire. Second edition. Pp. xi+422. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1959.) 56*s*. net.

THIS is a revised edition of a book originally published in 1947. The format and the heavy glossy paper are typically North American. The text is enriched by many photographs and diagrams; some of the photographs appear to have suffered in reproduction.

As Prof. Daubenmire's introductory definition of autecology in his preface shows, the sub-title of this book is misleading to British ecologists, most of whom would define autecology as the study of the ecology of a single species. This book, on the contrary, deals with the environmental factors which affect plants. Seven factors—soil, water, temperature, light, atmospheric, biotic, and fire—are given a chapter each, and two further chapters deal with "The Environmental Complex" and with "Ecologic Adaptation and Evolution". Much useful information on recent work is included, particularly in the fields of plant physiology and geneecology. Pursuing his interpretation of autecology, the author discusses the environmental factors as they affect particular plants, but the plant examples are chosen at random, and nowhere is there an integrated study of a single species such as a British ecologist would expect from "A Text-book of Plant Autecology". Nor is there any reference to the plant as part of a natural community, nor any analysis of different types of vegetation, nor any reference to the distribution of plants and historical ecology; presumably all these subjects fall within the author's definition of synecology and are deliberately excluded, but it is surely impossible to study the autecology of a species without at least considering these aspects. The approach to ecology presented in this book is therefore that of the experimentalist rather than that of the naturalist; indeed, "Plant Physiology in the Field" would be an apter title than "A Text-book of Plant Autecology".

Within these limits, the book will be of interest to plant geographers and should prove a useful reference book to students of botany. It provides data on many subjects which are relatively unfamiliar in Britain, such as frost action on soils, anatomical details of frost damage and sun scorch, anatomical effects on trees of the adverse habitats encountered at the upper timber-line of natural forest, and the ecological effects of widespread fires. Sections in the chapters on water factors and light factors provide a very useful bridge between plant physiology and ecology, and there is much miscellaneous but useful information in the chapters on atmospheric factors and biotic factors.

W. TUTIN

NON-INDIGENOUS ANIMALS OF GREAT BRITAIN

The Ark in Our Midst

The Story of the Introduced Animals of Britain—Birds, Beasts, Reptiles, Amphibians, Fishes. By R. S. R. Fitter. Pp. 320+16 plates. (London: William Collins, Sons and Co., Ltd., 1959.) 18*s*. net.

THIS is an ambitious book for in it the author sets out to cover a wide field, to survey the non-indigenous animals of the British Isles, that is the mammals, birds, reptiles and amphibians, which owe their inclusion in our fauna to the agency of man, creatures so diverse as the edible frog, the pheasant and the grey squirrel; and he does not restrict himself to those that have been successfully naturalized but includes species that have failed to establish themselves, pointing out that they are not the less interesting because they lack the necessary adaptability. Mr. Fitter tells us that the failures have been many and in excess of the successes, though the latter have often been spectacular with far-reaching results, for example, the manner in which the rabbit, the rat and the grey squirrel have colonized Great Britain.

Although we may be sure that man was the agent concerned in the arrival here of a number of creatures, although we may be positive they did not get here 'under their own steam', it is by no means easy to ascertain in each case how the deed was done, still less when it was done, even when it must have been well within the historic period.

It has long been the custom to give great credit to the Romans as importers of animals, much of which credit they undoubtedly deserve; yet evidence is singularly lacking as to what beasts, birds, fishes and so on they really did bring across the English Channel and naturalize in Britain. It is said that they introduced the pheasant; yet the bird bones found in Romano-British settlements have all been identified by experts as belonging to the domestic fowl. But we do find evidence that the pheasant was in Britain by Norman times. In a charter of 1098, the Bishop of Rochester assigned to that City among other things sixteen pheasants from certain Kentish manors, and in 1100 the Abbot of Malmesbury was given a royal licence to kill hares and pheasants, which latter we may presume were as naturalized and wild as the hares.

Mr. Fitter's industry in the collection of facts regarding the arrival in Britain of this variety of creatures has been immense. His lengthy volume is packed with information, the facts being clearly and concisely related, resulting in a most useful book that will be valuable for reference for many years to come.

FRANCES PITT

THE ZOOGEOGRAPHY OF WESTERN NORTH AMERICA

Zoogeography

Edited by Carl L. Hubbs. (A Symposium presented on August 26–27, 1957, at the Stanford University Joint Meeting of the American Institute of Biological Sciences and the Pacific Division of the American Association for the Advancement of Science, and a Symposium presented on December 28, 1957, at the Indianapolis Meeting of the American Association for the Advancement of Science.) Pp. x+509. (Publication No. 51.) (Washington, D.C.: American Association for the Advancement of Science; London: Bailey Bros. and Swinfen, Ltd., 1958.) 108*s*.

THIS volume contains seventeen full articles dealing with the wide variety of topics to be expected in two symposia on zoogeography. It does not deal much with the subject as a whole, and the title is really misleading, since most of the articles are concerned only or mainly with the western part of North America. The topics covered include the present and past distribution of land areas, habitats and climate (Tertiary and Pleistocene), physiological tolerance of vertebrates as bearing on distribution changes, Tertiary mammals and birds, Amphibia and reptiles, freshwater fish and invertebrates, Orthoptera, Dermaptera, longicorn beetles, and butterflies. The standard of scholarship is high, but inevitably for most of these groups more can be said of their present than of their past distribution, and we are constantly aware of the central difficulty of reconstructing faunal histories without the sort of fossil records that exist for the mammals, where the story can be best authenticated. Nevertheless, this volume is really valuable in drawing together into a coherent form a very large amount of information, and it represents much concentrated and intelligent research. As a great deal of the material upon which zoogeography is based inevitably accumulates in a rather haphazard manner, through the results of expeditions, museum bulletins, and so on, it is necessary to take stock at intervals and in this sort of way. It does not appear, however, that any very new ideas were brought to the surface by these symposia. This being so, it is a pity that the only index is one of Latin names. Anyone who wants to find other subjects will have to search the 477 pages of text.

To the general reader, perhaps the following four papers are the most useful. P. B. King traces the growth of modern surface features in Western North America, through the comparatively small deposition and mountain movements up to Jurassic times, and then the major upward Cordilleran movements and their aftermath. D. E. Savage gives a thorough conspectus of the fossil land mammals and their bearing upon Western faunal history. R. R. Miller has a very informative paper on the present distribution and origins of freshwater fishes of the West. A large paper on Pleistocene palæo-ecology by P. S. Martin covers a wider scene, including all North America, the West Indies and the Galapagos Islands, and takes in the arrival of man, and his influence on the fauna.

BUTTERFLIES AND MOTHS

Les Papillons

Par Guy Mathot. ("Que Sais-je ?" No. 797.) Pp. 128. (Paris: Presses Universitaires de France, 1958.) n.p.

"LES PAPILLONS" is not 'just another book on Lepidoptera'. There are very few books which aim at giving a popular account of world Lepidoptera and, in spite of the small size of this book (the page size is similar to the "Wayside and Woodland" series), it covers the subject very well.

There are six chapters: from egg to adult; morphology and anatomy of the imago; infra-specific variation; systematic studies on the principal groups of the world fauna; biogeography of the Lepidoptera; summary of biological studies.

The drawings in the book are not of a very high standard and the reproduction is poor, although this may be due to the poor quality of the paper. The text is very detailed in some parts, giving, for example, the chemical composition of various pigments, chromosome numbers and a table of wing loadings (p. 33). Some of the detail could have been left out in favour of other information. The table of wing loading (p. 33) and that (p. 43) giving the respiratory quotient seem unnecessary.

Small errors occur. The classification used (pp. 56–57) is not entirely satisfactory. The use of "Pyralidididoidea" and "Pyralididae" instead of the shorter Pyralidoidea and Pyralidae is irritating. A few spelling mistakes have occurred, *A. kühniella* Zell. is *kuehniella* (p. 81). The genus "*Paraponyx*" (p. 81) is always wrongly spelled and should be *Parapoynx* (Hubner, 1825). Some of the nomenclature is out of date (for example, *Lycaena* is used p. 52) instead of *Polyommatus* (Latreille, 1804).

The lack of authors' names after the species anywhere in the book is regrettable. Even in a book popularizing the subject the student should be correctly instructed, and in this case the author's name should have been given, at least where the species was first used.

In spite of these criticisms, the book contains a wealth of information unobtainable in any book of comparative size. It will interest all collectors who wish to study more than just British Lepidoptera.

PAUL E. S. WHALLEY

The Freshwater Life of the British Isles

A Guide to the Plants and Invertebrates of Ponds, Lakes, Streams and Rivers, with an additional chapter on the Vertebrates. By John Clegg. Second edition. (Wayside and Woodland Series.) Pp. 352 + 67 plates. (London and New York: Frederick Warne and Co., Ltd., 1959.) 21*s*. net.

THIS popular book, first published in 1952, deals in broad outline with the basic information necessary to an understanding of limnology; it has proved its worth as an elementary text for teachers and students and, in particular, for those attending courses at the field study centres.

This new edition has given Mr. Clegg the opportunity to eliminate some errors which had crept into the first edition and to include some new results that had become available since 1952. Some colour plates by Mr. Ernest C. Mansell are noteworthy additions, those on Mollusca being particularly good. The opportunity might also have been taken to provide more satisfactory colour photographs both of the natterjack toad and of the common toad.

Although a great many animals and plants can be identified with the aid of this book, there are no keys, and it is not primarily concerned with identification down to species. Illustrated extended keys (of too recent date to have been included in the revised bibliography) are now available and can be used in conjunction with this volume, but they do not pretend to provide a comprehensive, well-balanced introduction to limnology such as the beginner will find in Mr. Clegg's book. W. J. Rees

The Future of Non-Ferrous Mining in Great Britain and Ireland

(Proceedings of a Symposium arranged by the Institution of Mining and Metallurgy in collaboration with the United Kingdom Metal Mining Association, held on 23rd and 24th September, 1958, at the Federation of British Industries, London.) Pp. xxxvi + 614. (London: The Institution of Mining and Metallurgy, 1959.) 60*s*.

THE main scientific results of the symposium organized jointly by the Institute of Mining and Metallurgy and the United Kingdom Metal Mining Association have already been reported briefly in *Nature* (Oct. 25, 1958, p. 1136). Publication of the Proceedings, delayed by the printers' strike, has now been achieved. The single volume contains the text of the fourth Sir Julius Wernher Lecture, delivered as a prelude to the symposium by Mr. Anton Grey. This is followed by thirty-two original papers, most of which had been issued as pre-prints to members, together with verbatim reports of the discussions at the two days of meetings. Sixteen of the papers are devoted to descriptions of the tin, copper, lead, zinc, fluorine and barium deposits of the British Islands, the most comprehensive review of the epigenetic mineral deposits of Britain so far assembled in a single volume. History, geology, production and potential are dealt with in each of these papers. Most of the ore-fields have been described before, some of them in some detail, as for example in the Geological Survey Memoirs of the Northern Pennine Ore-field (1948) and South-Western England (1956), some only in a general fashion. The present publication gives an up-to-date picture of an industry which is now no more than a shadow of its former self. The purpose of the symposium was to consider whether resources of the non-ferrous minerals remain in the British Isles after more than 2,000 years of exploitation; whether these are worth finding, and if so how they can be located. With these points in view, the other papers and discussions were devoted to modern techniques for prospecting—geophysical, geochemical, radiometrical—and to considerations of economics and ownership. If the conclusions can be summed up in a single phrase, they are that deposits as yet undiscovered may be expected to remain; that in all but a few cases the search for these would be so speculative as to need some form of State support; and that when they are found the value of the deposits against the current economic background cannot be said to be assured.

K. C. Dunham

Infant Mortality in India, 1901–55

A Matter of Life and Death. By Prof. S. Chandrasekhar. Pp. 175. (London: George Allen and Unwin, Ltd., 1959.) 20*s*. net.

IN writing this book Prof. Chandrasekhar essays the difficult task of being three persons at once. As director of the Indian Institute for Population Studies he makes clear the inadequacy of available statistics. Though the wealth of material in the decennial censuses of India since 1881 is well known, essential basic data such as total number of births, or total number lost under one year, are not available. In the second place the author, as a distinguished social scientist, has little difficulty in showing how unfavourably India compares with so many other countries. Though the infant mortality-rate is shown as having dropped from 215 per 1,000 live births in 1901–05, to 116 in 1951, the latter figure is compared with such examples as 29 in the United States or 27 in England and Wales. In fairness it is noted that infant mortality is still believed to average 300–500 per 1,000 live births over most of Africa. In the third part of the book the author, as social reformer, claims that the nation's children are too precious to be wasted by premature death, that the nation's efforts should be directed to the ideal of reducing the present infant mortality-rate to something like 20 per 1,000 live births. Family planning with a limitation to, "say, three under normal circumstances spaced, say, three years apart," is mentioned incidentally rather than made a focal point.

L. Dudley Stamp

Second Conference on Co-ordination of Galactic Research

Edited by A. Blaauw, G. Larsson-Leander, N. G. Roman, A. Sandage, H. F. Weaver and A. D. Thackeray. (International Astronomical Union Symposium No. 7, held at Saltsjöbaden, near Stockholm, June 1957.) Pp. viii + 93. (Cambridge: At the University Press, 1959.) 15*s*. net.

THE first conference on the co-ordination of galactic research was held in 1953 and its report appeared as International Astronomical Union Symposium No. 1; it was devoted to a broad survey of the problems of galactic structure. The second conference was devoted to more detailed problems of particular importance at this time: clusters and associations, the Galaxy as a whole, and the Magellanic Clouds. A good account is given of many aspects of current work as well as discussions of the possibilities for future investigations. Emphasis is laid on the importance of adequate standardization of the photometric systems used by different observers.

ROLE OF INFECTIVE NUCLEIC ACID IN THE PRODUCTION OF ENCEPHALOMYOCARDITIS VIRUS

By Dr. F. K. SANDERS

Director, Virus Research Group, Medical Research Council Laboratories, Carshalton, Surrey

COLD phenol treatment[1] of cells infected by several different animal viruses yields infective material, a major constituent of which appears to be ribonucleic acid[2–7]; in this article such preparations will be referred to as 'infective ribonucleic acid'.

In some cases, infective ribonucleic acid seems to come from the virus itself; but in others to form part of another component, which does not sediment appreciably under conditions which bring down virus[6,7]; in the latter cases phenol at 4° C. fails to extract infective ribonucleic acid from the virus particles themselves, although hot phenol (40° C.–60° C.) may be able to do so[7]. The extracellular fluid into which virus has been released at the end of virus growth in mouse ascites tumour cells infected *in vitro* with murine encephalomyocarditis virus contains a component, sensitive to cold phenol, which is not the virus itself[6]. Such a component might be: (*a*) some kind of cellular structure formed as a result of infection, in relation to which virus is synthesized; (*b*) a precursor of the virus particle itself; (*c*) a product of the abnormal synthesis, which somehow fails to be incorporated into a virus particle; (*d*) a breakdown product of virus particles.

Continuously agitated suspensions of Krebs 2 mouse ascites carcinoma cells, infected with encephalomyocarditis virus *in vitro*[8], were used to follow the appearance of infective ribonucleic acid during virus growth, extracts being made from cells at all stages of the virus growth-cycle up to the beginning of virus release. Infected cells, washed with phosphate-buffered saline, and resuspended at a concentration of 5 × 10^7/ml. in 0·02 M phosphate pH 7·2, were extracted three times with water-saturated phenol. Phenol was removed from the aqueous layer with ether and the ribonucleic acid precipitated by ethanol. The precipitate was washed three times with 67 per cent ethanol/33 per cent 0·14 M sodium chloride and redissolved in 0·2 M phosphate buffer pH 6·8. All operations were carried out at 4° C. and all aqueous solutions except the 0·2 M phosphate buffer contained nickel ions to stabilize the ribonucleic acid; the optimal concentration of nickel was found to be 1 nickel ion per nucleotide, as already found for tobacco mosaic virus ribonucleic acid[9]. A small residual precipitate of nickel phosphate was removed by centrifugation. Ribonucleic acid preparations were mixed with tumour cells and titrated in mice[6], on the day of preparation, although the above method gave a product which retained its original titre for at least 14 days at 4° C., and 24 hr. at 20° C. (Vizoso, A. D., and Burness, A. T. H., personal communication).

In multiply infected cell populations (cells 1 × 10^7/ml.; virus 3 × 10^7/ml.) at 37° C. infective ribonucleic acid could be found as early as 15 min. after infection. Thereafter the level of infectivity rose to a maximum at about 5 hr., when 100–1,000 times the amount present initially was found. Infective ribonucleic acid production is thus practically complete by the time new virus is formed; the latter appears intracellularly at about 4 hr., and then increases to maximum titre 7–8 hr. after infection. Cells begin to die and release this virus 6–7 hr. after infection, and release from the whole population is virtually complete at 12 hr. The extracellular fluid then contains more than 2 × 10^9 plaque-forming units of virus/ml. and a similar number of mouse ID_{50} when titrated *in vivo*.

The increase in infective ribonucleic acid content found during the first hour probably does not involve synthesis of new ribonucleic acid, but represents the formation of an increasing number of virus/cell complexes, in which the ribonucleic acid of the infecting virus becomes capable of extraction by phenol, for cells infected for 2–16 hr. at 4° C. all contain ribonucleic acid with an infectivity corresponding to the level reached in 1 hr. at 37° C., and this level of infectivity is also reached in the presence of inhibitors (see below). Similarly, a fall of about 90 per cent in the infective ribonucleic acid content occurring between 5 and 7 hr. after infection (Fig. 1) need not involve the destruction of infective ribonucleic acid but its progressive inclusion in virus particles whence it cannot be extracted by cold phenol in an infective form.

Effect of Euflavine

Experiments in which virus was grown in the presence of euflavine gave further information about the relationship of the early appearance of infective ribonucleic acid to virus synthesis. In an initial experiment cells (10^7/ml.) were incubated for 1 hr. in the presence of various concentrations of euflavine,

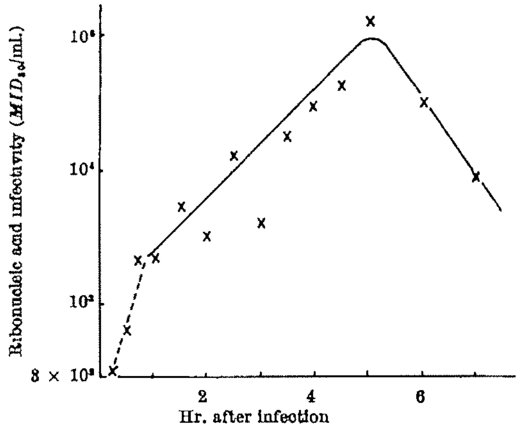

Fig. 1. Infectivity of 'cold phenol ribonucleic acid' at intervals during virus growth

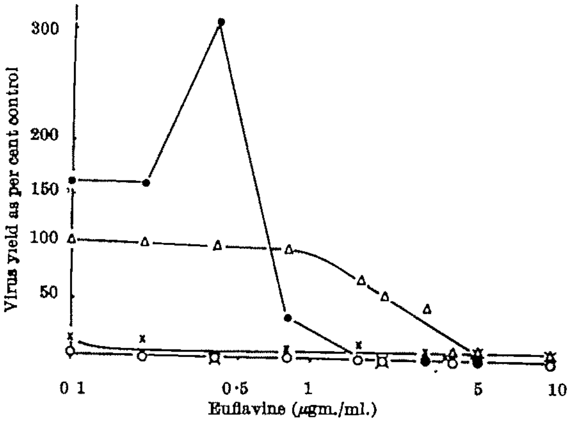

Fig. 2. Effect of various concentrations of euflavine on virus production. ○, plaque-forming units; ●, hæmagglutinin; ×, infective ribonucleic acid; △, cell mortality

all of which were without significant toxic effect upon control uninfected cultures in experiments of similar duration. Virus was then added (multiplicity = 4), and the culture incubated for 16 hr., when the following quantities were measured; (*a*) virus hæmagglutinin concentration, and (*b*) concentration of infective virus (plaque-forming units), in the supernatant fluid; (*c*) the infectivity of ribonucleic acid extractable from the whole culture (cells plus supernatant); (*d*) the number of cells killed by virus multiplication[6,8,10].

All euflavine concentrations tested, even the smallest (0·1 µgm./ml.) virtually suppressed the production of infective virus (Fig. 2); hæmagglutinin production was also inhibited by euflavine concentrations greater than 2 µgm./ml. However, in low concentrations (0·1–0·5 µgm./ml.), hæmagglutinin production was high, probably somewhat increased with respect to the control. Proflavine, a related drug, interferes with a late stage of bacteriophage multiplication—probably that of virus assembly[11,12]—and it is possible that low concentrations of euflavine can similarly disturb the multiplication of encephalomyocarditis virus. Even small amounts of euflavine significantly reduced infective nucleic acid formation, although high doses of the drug failed to reduce the infective ribonucleic acid content below that found in untreated cultures during the first hour after infection. With less than 1 µgm. euflavine/ml. as many cells eventually died as in the control, but in higher concentrations more and more cells appeared to resist the lethal effect of the virus, so that at concentrations greater than 4 µgm./ml., virtually all cells were preserved.

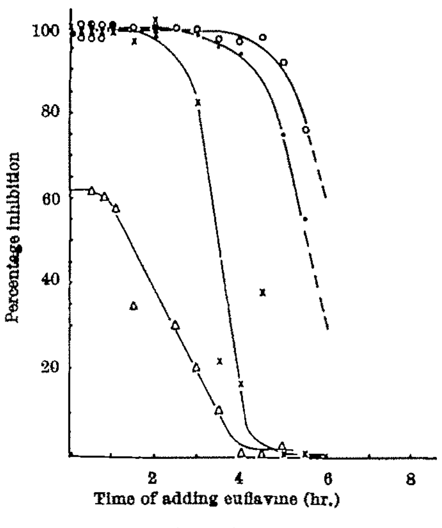

Fig. 3. Effect of adding 5 µgm./ml. euflavine at different times during virus growth. Symbols as in Fig. 2

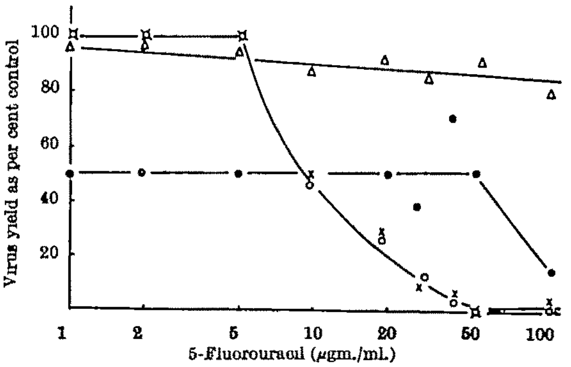

Fig. 4. Effect of various concentrations of 5-fluorouracil on virus production. Symbols as in Fig. 2

Fig. 3 gives the result of an experiment in which 5 µgm./ml. of euflavine was added to ascites tumour cell suspensions at different times after infection. Up to 1½ hr. this dose of euflavine prevented the production of all virus-related substances measured. When given at 4 hr., however, while the production of hæmagglutinin and plaque-forming units was still suppressed, the production of infective ribonucleic acid was normal and the cells died at the proper time. When euflavine was given later than 4 hr., more and more hæmagglutinin was found although even at 5½ hr. after infection there was still significant inhibition of the production of complete virus particles (plaque-forming units). Thus the production of infective ribonucleic acid must be virtually complete by the time hæmagglutinin begins to appear; the latter appears only to be incorporated into plaque-forming virus particles towards the end of virus growth.

Effect of 5-Fluorouracil

This pyrimidine analogue can become incorporated into the ribonucleic acids of animal tissues[13], bacteria[14] and tobacco mosaic virus[15] and it was thus of interest to discover what effect it might have on encephalomyocarditis infective nucleic acid and virus production; all concentrations tested were without significant toxic effect upon the cells under the conditions used. Doses of 5-fluorouracil up to 20 µgm./ml. had little effect on the production of hæmagglutinin and failed to prevent virus-infected cells from dying at the end of the virus growth-cycle (Fig. 4). However, with doses greater than 5 µgm./ml. there was a progressive diminution in the number of plaque-forming units, so that at 50 µgm./ml. or more, less than 1 per cent of the normal number was found. High levels of the drug also affected the production of hæmagglutinin although to a smaller extent. The curve of suppression of infective ribonucleic acid production was very similar to that for plaque-forming units, which suggested that infective ribonucleic acid might actually become incorporated into the virus particle. In further experiments 100 µgm./ml. 5-fluorouracil was added at various times after infection (Fig. 5).

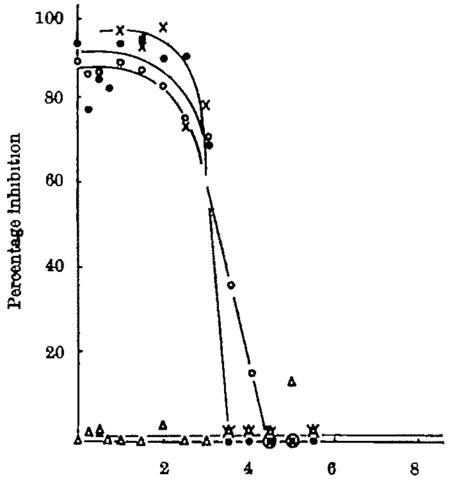

Fig. 5 Effect of adding 100 μgm./ml. 5-fluorouracil at different times during virus growth. Symbols as in Fig. 2

When this amount of 5-fluorouracil was given 1 hr. before virus, there was marked inhibition of hæmagglutinin production and the production of infective ribonucleic acid and plaque-forming units was suppressed; but there was no sparing of the cells. Up to 1 hr. after infection, its effect was the same. With later administration of the drug, however, more infective ribonucleic acid, more hæmagglutinin and more infective virus particles were produced. Addition of the drug at any time after 4 hr. failed to affect the course of virus multiplication. 5-fluorouracil presumably acts during nucleic acid synthesis by replacing part of the uridylic acid residues with their 5-fluoro analogue[14,15]. Anomalous nucleic acid containing this substance is presumably not infectious, and virus particles with such a nucleic acid would not be able to reproduce themselves. The fact that the time-course of suppression by 5-fluorouracil is the same for both these entities suggests that the ribonucleic acid formed between 1 and 4 hr. after infection eventually becomes part of the virus particles. Moreover, since hæmagglutinin production is only significantly affected by 5-fluorouracil when the drug is given during infective ribonucleic acid formation suggests that the newly synthesized ribonucleic acid may act as a centre for the later synthesis of hæmagglutinin.

The multiplication of encephalomyocarditis virus thus probably takes place as follows. Contact of a virus particle with its host cell causes the formation of a virus/cell complex; this can be defined operationally as some alteration in the virus structure resulting from contact with the cell which releases the virus ribonucleic acid in a form extractable by phenol at 4° C. Complex formation presumably involves neither nucleic acid nor protein synthesis since it is not inhibited by either fluorouracil or euflavine, and takes place easily at 4° C. and at 37° C. The next stage seems to be replication of ribonucleic acid carrying the genetic information of the virus, which probably begins at about one hour after infection and reaches a maximum about four hours later; it gives rise to the component described earlier[6], which is the source of the infective ribonucleic acid extracted by cold phenol from infected cells. Only later comes the production of virus hæmagglutinin from 3½ to 6 hr. after infection, and this stage can still be inhibited by euflavine added 4 hr. after infection, though not by 5-fluorouracil added at this time. In the final stage of virus reproduction a core containing the nucleic acid and the hæmagglutinin are put together to make a virus particle. This phase can be disturbed by concentrations of euflavine which are too small to affect hæmagglutinin production. The ribonucleic acid component thus seems essentially to be a virus precursor. Besides its possible function as a centre for the later synthesis of hæmagglutinin, it may eventually become incorporated into the virus particle itself. The process of incorporation is not, however, 100 per cent efficient and a proportion of the component fails to get included in virus particles; it is this extra material which gives rise to the infective ribonucleic acid that can be extracted from the extracellular fluid at the end of virus growth.

It has been stated elsewhere[8] that infection of a cell by virus starts two processes—one resulting in the production of progeny virus, and the other in cell death. The train of events which leads to death of the cell seems to depend on some key change which takes place very early in the growth-cycle. Procedures which favour the production of virus containing anomalous nucleic acid, interfere with the synthesis of hæmagglutinin, or the assembly of virus particles, do not normally prevent cell destruction at the proper time. Blocking virus synthesis by adding 5 μgm./ml. euflavine within ½–1 hr. of infection preserves the cells from eventual destruction, which suggests that the irrevocable event which eventually leads to cell death may be the start of synthesis of the ribonucleic acid component itself. However, evidence from analysis of the interaction between encephalomyocarditis virus strains of narrow host-range, and ascites cells with which they are unable to form plaques, indicates that this may not be so. Cells of the Ehrlich carcinoma or sarcoma 37 are able to take up encephalomyocarditis virus of mouse brain origin[8,16], although new hæmagglutinin or plaque-forming virus is not produced as a result of infection. Large amounts of infective ribonucleic acid—as much as is produced by the fully susceptible Krebs 2 cells—can be extracted from such abortively infected Ehrlich or sarcoma 37 cells, and a component containing this ribonucleic acid is set free into the external medium. Such cells do not die as a result of infection, so death of a cell need not be an inevitable consequence of synthesis of viral ribonucleic acid.

[1] Schuster, H., Schramm, G., and Zillig, W., *Z. Naturforsch.*, **11**, *b*, 339 (1956).

[2] Colter, J. S., Bird, H. H., Mayer, A. W., and Brown, R. A., *Virology*, **4**, 522 (1957).

[3] Alexander, H. E., Koch, G., Mountain, I. M., and Van Damme, O., *J. Exp. Med.*, **108**, 493 (1958).

[4] Wecker, E., and Schafer, W., *Z. Naturforsch.*, **12**, *b*, 415 (1957).

[5] Franklin, R. M., Wecker, E., and Henry, C., *Virology*, **7**, 220 (1959).

[6] Huppert, J., and Sanders, F. K., *Nature*, **182**, 515 (1958).

[7] Schafer, W., *Symp. Soc. Gen. Biol.*, **9**, 61 (1959).

[8] Sanders, F. K., Huppert, J., and Hoskins, J. M., *Symp. Soc. Exp. Biol.*, **12**, 123 (1958).

[9] Cheo, P. C., Friesen, B. S., and Sinsheimer, R. L., *Proc. U.S. Nat. Acad. Sci.*, **45**, 305 (1959).

[10] Sanders, F. K., *Proc. Roy. Soc. Med.*, **50**, 911 (1957).

[11] Foster, R. A. C., *J. Bacteriol.*, **56**, 795 (1948).

[12] Kellenberger, G., and Kellenberger, E., *Virology*, **3**, 275 (1957).

[13] Chaudhuri, N. K., Montag, B. J., and Heidelberger, C., *Cancer Res.*, **18**, 318 (1958).

[14] Horowitz, J., and Chargaff, E., *Nature*, **184**, 1213 (1959).

[15] Gordon, M. P., and Staehelin, M., *Biophys. Biochim. Acta*, **36**, 351 (1959).

[16] Hoskins, J. M., *Symp. Soc. Gen. Microbiol.*, **9**, 122 (1959).

FALL-OUT OVER NORWAY FROM HIGH-YIELD NUCLEAR EXPLOSIONS

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FROM measurements made in Norway during the past three to four years it seems that fall-out materials which rise well into the stratosphere after high-yield test explosions begin to reach the lower stratosphere over south-east Norway shortly afterwards. The radioactivity in the lower stratosphere seems to increase to a peak about one to two months after the explosions and then to decrease with an effective half-life of about a month. This is shown in Fig. 1, curve *A*, the arrows indicating the delay times. The radioactivity of fall-out materials in air near the ground also begins to increase shortly after the explosions, but more slowly, and seems to reach a peak, which is considerably lower than the peak in the lower stratosphere, after about six to seven months. This is shown in Fig. 1, curve *B*, the delay times being once again indicated by arrows. The rate of increase apparently results in a doubling of the radioactivity in the course of a month or so. The subsequent rate of decrease appears to be the same as in the stratosphere, the radioactivity being halved about once every month. The radioactivity of fall-out materials in precipitation begins to increase at about the same time as the radioactivity of air near ground-level, and follows very much the same pattern. This is shown in Fig. 1, curve *C*.

An interesting observation from Fig. 1 is that the rates of decrease of radioactivity in the atmosphere after large additions of fall-out materials during the years 1957–59 have been remarkably alike, regardless of the overall amounts of fall-out materials in suspension. By correcting for radioactive decay, it can be calculated that the effective residence half-life of fall-out materials in the lower stratosphere over south-east Norway has been of the order of three months.

Since fall-out materials may begin to arrive in air near ground-level shortly after high-yield explosions, it follows that during periods when explosions occur at relatively short intervals there may be considerable mixing of materials near the ground. This can be illustrated by the position at the end of

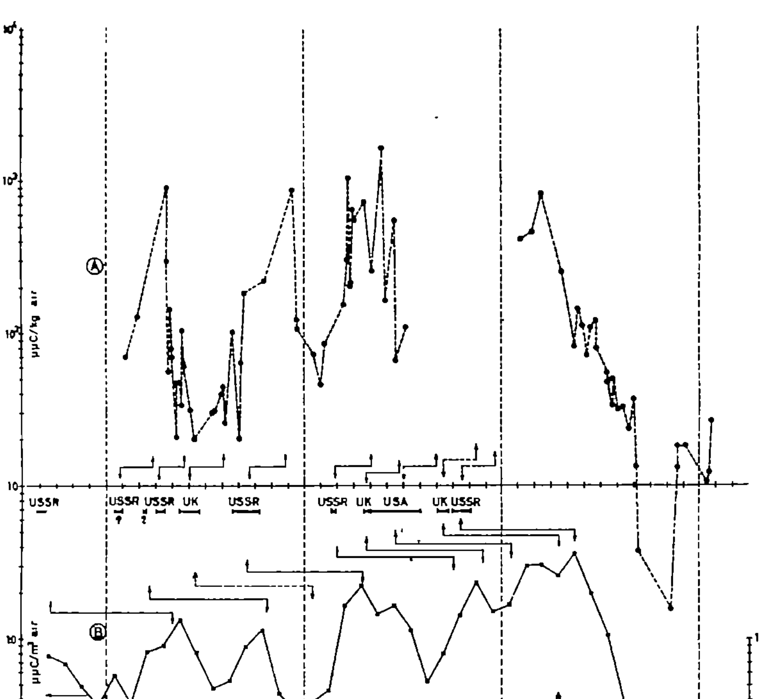

Fig. 1. Results of measurements of fall-out materials during the period September 1956–January 1960

Curve *A*, concentrations of fall-out materials in the lower stratosphere over south-east Norway. Each point represents the result of one measurement. Curve *B*, monthly average concentrations of fall-out materials in air near ground-level. Each point represents an average value of measurements from nine stations. Curve *C*, monthly average concentrations of fall-out materials in precipitation. Each point represents an average value of measurements from twelve stations

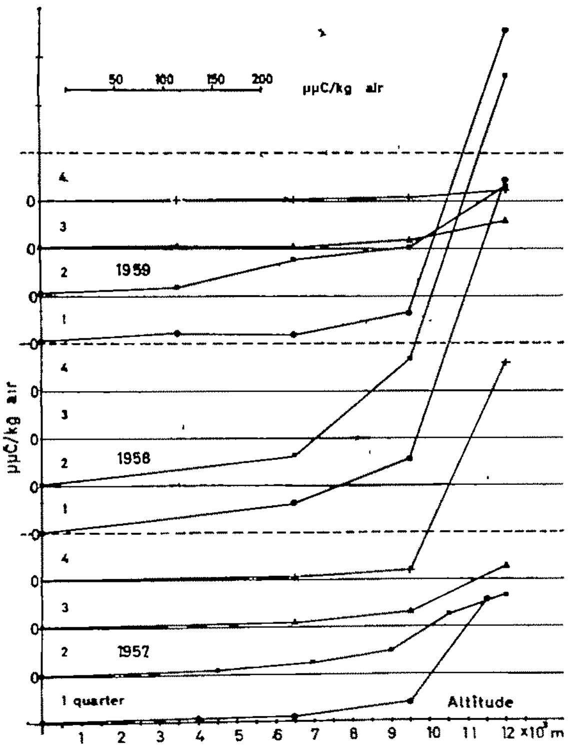

Fig. 2. Concentrations of fall-out materials at different heights above ground-level over south-east Norway during 1957–59. Most points represent the average of 5–10 measurements made during each quarter

1958, when materials from at least five test explosion series could have been present in significant quantities. It is therefore problematical whether isotope ratios or decay-rates can provide reliable information about the origins of fall-out materials at such times. There is no outstanding seasonal variation in Fig. 1 which cannot be explained by the 'seasonal variation' in test explosions.

It has been found that the concentrations of fall-out materials in precipitation depend to some extent upon amounts of precipitation, which vary very much. Consequently it is not to be expected that curves *B* and *C* of Fig. 1 should be completely alike. Nevertheless, there is a very obvious similarity between them, and it can be deduced that on the average 1 cm.^{3} of precipitation has contained about the same amount of fall-out materials as 1 m.^{3} of air near ground-level.

The curves shown in Fig. 2, which indicate the way in which concentrations of fall-out materials vary with height above sea-level, might at first sight be interpreted as indicating that downward transport of fall-out materials is hindered by the tropopause layer. However, it seems very possible that the marked increase in concentrations at about tropopause height is due to the rapid removal of materials from lower air layers. Fig. 2 would then indicate that removal processes are effective up to heights of several km. By comparing measurements of airborne materials with measurements of deposited materials, it has been calculated that it would have been necessary to remove the total content of fall-out materials in the troposphere over south-east Norway about once every month, in order to account for the measured deposition. This implies a rather rapid rate of supply to the troposphere, and is interesting in connexion with the short residence half-life of fall-out materials in the lower stratosphere deduced from Fig. 1.

Fig. 3 shows that the main variations in concentrations of fall-out materials in surface air occur approximately simultaneously at measurement sites from Vadsø in the north to Sola in the south, a distance of about 1,700 km. This is quite interesting because the sites are at different heights above sea-level, and are exposed to markedly different weather conditions. It seems clear from Fig. 3 that if fall-out materials are mainly removed from the stratosphere by downward air movements, then these must occur rather consistently over a wide region.

Although much more work must be done to find out what happens to fall-out materials after a nuclear explosion, it now seems that deposition is a fairly continuous short-term process, lasting months instead of years, and affecting very large areas. It is possible that fall-out particles may achieve significant settling-rates, particularly in the lower stratosphere and upper regions of the troposphere. Removal from the stratosphere by downward air movements would then be relatively less important than previously assumed.

In view of the short deposition times, measurements of external radiation-levels and gross activity

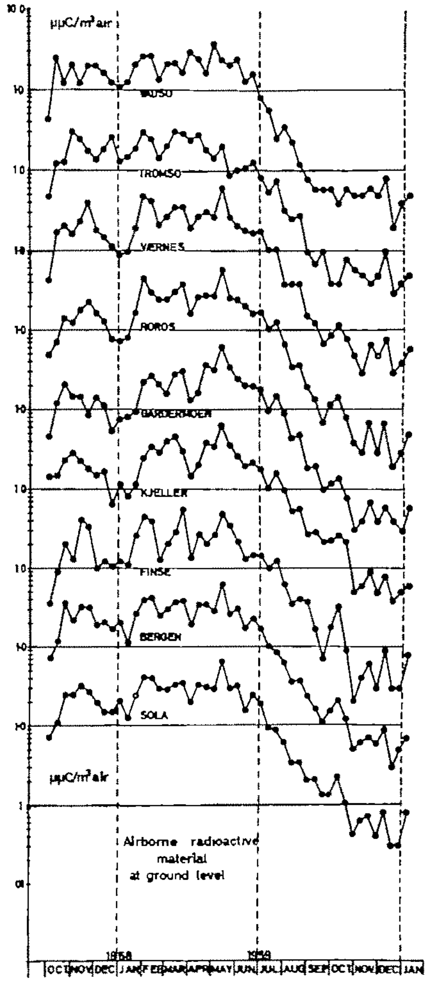

Fig. 3. Mean measurements over ten-day periods of concentrations of fall-out materials in air near ground-level at different stations in Norway during October 1958–January 1960

measurements now have an increased importance, in relation to measurements of selected long-lived isotopes, in assessing possible radiation hazards.

The investigations described in this article form part of a programme of fall-out measurements carried out by the Norwegian Defence Research Establishment. We wish to acknowledge the help given by the Royal Norwegian Air Force and the Norwegian Meteorological Service in collecting samples of fall-out materials, and by members of the Physics Division of the Norwegian Defence Research Establishment in measuring samples, etc.

RADIOACTIVE MATERIALS AND THE SKELETON

THE one-day conference organized by the Bone and Tooth Society on "Radioactive Materials and the Skeleton" included papers on tracer applications of bone-seeking isotopes in biology and in medicine, studies of their biological effects, measurements on rates of deposition of strontium-90 in fall-out, the entry of strontium-90 into food, a survey of the radiation doses received by man from environmental sources and some discussion on the biological implications of environmental radiation in so far as bone was concerned. It was, in fact, a very ambitious programme for a short meeting, but it served the very useful purpose of allowing this controversial subject to be seen as a whole.

The first paper, "Anatomical Sites of Deposition of Radioactive Materials in Bone", was given by Prof. P. Lacroix, of the University of Louvain, who has himself been responsible for many fundamental advances in the field of the dynamics of bone growth. He showed calcium-45 autoradiographs of bones of young and adult dogs and discussed the relevance of the variations in the uptake of calcium throughout the bone to our understanding of the various processes of bone growth and remodelling which take place. Of particular interest was the change in the pattern of uptake with age of the young animal. Soon after birth the major mode of transverse growth of the long bones is by the peripheral deposition of thick layers of bone, the structure of which is still undetermined. This, however, gives way to transverse growth mainly by a process of deposition of rows of osteones under the periosteum, with a different pattern of calcium uptake, which is clearly demonstrated by autoradiography.

In clinical work with bone-seeking isotopes the material for autoradiographic investigation is generally not available and the distribution of the administered isotope must often be determined by external counting methods, which give relatively poor resolution. Much less information is derived than from experimental biological studies but still, as the second paper of the conference, "Bone-seeking Isotopes as Indicators of Bone Turnover", by Drs. G. Bauer and Bo Wendeberg, of Malmö, showed, information can be obtained on the turnover of bone mineral in pathological conditions of bone which cannot be obtained by other methods.

A study of the metabolism of strontium, with strontium-85, in human subjects using two volunteers, was next described by Dr. G. E. Harrison, of the Medical Research Council Radiobiological Research Unit, Harwell, in his paper, "The Skeletal Retention of Radioactive Strontium in Man following an Intravenous Dose". Although a dose of only 0·5 μc. strontium-85 was given to each subject, it was possible to follow the retention pattern for one year, first using excretion data and then measurements on retained activity by whole-body counting. The results for the two subjects were remarkably similar, demonstrating the early initial excretion and then a period of relatively rapid loss with a half-life of about 50 days followed by a slow loss from the relatively unexchangeable part of bone. The calculated mean radiation dose to the skeleton over a period of 300 days was 0·23 rad.

The three papers of the second session dealt with the biological effects of bone-seeking radioactive isotopes. There is a considerable literature on this subject relating mainly to rates of tumour incidence with different isotopes. Large differences in tumour incidence have been found with different isotopes and different patterns of administration ; but the significance of such differences are obscure without information on the distribution of radiation dose in bone and its variation with time, and such information is generally not available.

The first two papers both discussed the histological changes in the bones of growing animals in relation to measured radiation dosage distribution. The first paper, "The Relation of Radiation Damage to Radiation Dose from Strontium-90", by Drs. Sheila Macpherson and Maureen Owen, of the Medical Research Council Group for Research on Bone-Seeking Isotopes, Oxford, dealt with the effect of a long-lived isotope in rabbits, whereas the second, "Bone Tumour Production : Dose Response Relationships using Phosphorus-32", by Drs. J. P. M. Bensted and N. M. Blackett, of the Physics Department, Institute of Cancer Research, London, discussed the effects of a relatively short-lived isotope in rats. In spite of these differences there was much similarity in the gross biological effects observed and both papers demonstrated the considerable reparative capacities of bone to large radiation doses, of the order of thousands or even tens of thousands of rads. With regard to possible histological antecedents of bone tumour production the two sets of experiments had less in common. Whereas with repeated doses of phosphorus-32, which is a particularly efficient method of producing bone tumours in the rat, an increase in connective tissue in the metaphysis appeared to be closely related to bone tumour production, the experiments with strontium-90 did not demonstrate this. In both sets of experiments the tumours observed arose after very heavy radiation doses, of the order of thousands of rads.

These two papers pointed to the work that remains to be done in this field, rather than to what has been achieved. Certainly the mechanisms of bone tumour production by radiation and the likelihood of bone tumours appearing at low dosage-levels are not yet subjects for easy speculation.

Abnormality in tooth development of young animals given large doses of bone-seeking isotopes is a common finding in experimental work, but relatively little work has been done on the histological changes involved. Prof. M. A. Rushton, of Guy's Hospital Medical School, in his paper, "Damage to Dental Tissues in High-Dose Rabbit Experiments", presented a detailed histological study of the response of the developing teeth of weanling rabbits given large doses of strontium-90 (600–1,000 μc./kgm.). The damage observed was very severe; but the radiation doses were extremely high, of the order of tens of thousands of rads. It is greatly to be hoped that work of this calibre can be extended to much lower levels of radiation dose.

The third session was devoted to problems of environmental radioactivity where the radiation dose-rates involved are of the order of a fraction of a rad a year. The first two papers presented dealt with the measurement, and some aspects of the metabolism, of strontium-90. In order to measure the amounts of strontium-90 deposited in Great Britain from fall-out and the resulting labelling of our foodstuffs a considerable organization has had to be built up, whereby samples can be collected from any parts of the country. In their paper, "Strontium-90 in the Environment and in Human Bone", Drs. F. J. Bryant and D. H. Pierson, of Atomic Energy Research Establishment, Harwell, described measurements of strontium-90 content in rain, in drinking water, in human bone and teeth. Apart from the number of test explosions made there are two main factors determining the rate of deposition of strontium-90. The first is the magnitude of the explosions. Debris from explosions of kiloton bombs does not penetrate the tropopause and has a mean residence time in the troposphere of about 30 days. The debris from megaton bombs is, however, injected into the stratosphere and the mean residence time may be as long as 10 years. The second determining factor is the total rainfall over the given period. The strontium-90 content in tap-waters derived from lakes in the United Kingdom in early 1959 was 0·9 μμc./l., with lower values for river waters and wells. The highest strontium-90 content of human bones was found in the age-group 0–5 years and had the average value in the femur of 0·83 μμc./gm. in the second half of 1958.

Dr. R. Scott Russell, of the Agricultural Research Council Radiobiology Laboratory, Wantage, in his paper, "Radioactive Materials in Foodstuffs", discussed the uptake of strontium-90 in plants and vegetation. He stressed that the major part of the strontium-90 contamination of our foodstuffs came from direct deposition of strontium-90 on to the plant rather than via the soil. It could also be shown that there was only a short interval, not more than about two months, between the deposition of strontium-90 and its appearance in milk, the major source of calcium in our diet in Great Britain. Consequently the strontium-90 content of our food was much more dependent on the rate of deposition of fall-out than on the total accumulated amount.

It must have been very clear from these two papers that the information so far obtained on the deposition of strontium-90 in fall-out and its progress through the food chain is the result of a concentration of highly skilled and intelligent effort and has, in the course of the work, opened up new lines of advance in many fields, from meteorology to agriculture. However, the object of the exercise was to learn something of the biological consequences of fall-out, and the measurement of levels of activity can represent only the preliminary step. The translation of these measured quantities into biological effect is the real challenge.

The values of radiation dose-rate in bone resulting from present levels of fall-out was included in the paper given by Prof. F. W. Spiers, of the University of Leeds, on "Radiation Doses to Bone from all Sources". He first dealt with the problem of where in bone one should measure the radiation dose. Clearly the choice must depend on the degree to which the various tissues comprising bone are significant in the development of late effects. This, however, is not known and the significant dosage values must therefore be uncertain, particularly for the alpha-emitting isotopes, on account of their short range. For strontium-90, with its relatively long beta range, there is still a problem, though not so severe as with the alpha emitters.

Prof. Spiers presented tables of the radiation doses received by bone and by soft tissue from cosmic rays and local gamma-rays, from the natural radioactivity of tissues, and from the strontium-90 and cæsium-137 originating in fall-out. Within Great Britain the total dose-rate received from all sources is of the order of 100 millirem per annum, and of this amount cosmic radiation and local environmental radioactivity contribute about 90 per cent. At present levels the strontium-90 contribution to bone does not normally exceed about 4 millirem a year. The contribution to gonadal tissue is extremely small. The major variation in environmental radiation dose received by an individual depends on the gamma-ray contribution from the local environment, but variation in the natural alpha activity of tissues also plays an appreciable part. As a result of these variations in natural radioactivity the dose-rate in bone may vary between individuals by a factor of as much as 2.

Radiation dosage data such as those presented by Prof. Spiers allow one to come to certain conclusions concerning the comparative somatic effects of the various components of environmental radiation. It is difficult, for example, to avoid the conclusion that if present levels of strontium-90 in bone constitute a hazard, a change in locality may incur a much greater hazard. Beyond such comparative statements it is difficult to go, since the biological data relevant to the late radiation changes in bone refer to radiation dose-levels several orders of magnitude higher than that of environmental radiation. Any theoretical treatment of the problem of the somatic effects of small doses involves the widest assumptions—on the part played by specific cell changes, on the significance of gross tissue damage, on the types of tissue at risk and on the part played by the normal homœostatic mechanisms of the body. Nor is it at all certain that a direct attack on the problem, by giving small doses of radiation to large numbers of experimental animals, is justified. In my paper, "The Biological Implications of Radiation Dosage to Bone from Environmental Radioactive Material", I suggested that too much preoccupation with the quantitative aspects of the late effects of low-level irradiation could, in the present state of knowledge, be a great waste of effort and even a hindrance to the better understanding of the subject. It would seem more reasonable to accept the fact of our present ignorance, to rely on comparative radiation dosage data for necessary action and to concentrate experimental effort on a study of the mechanisms, rather than the statistical aspects, of late radiation effects.

L. F. LAMERTON

OBITUARIES

Sir Ernest Barker, F.B.A.

THE death on February 17 of Sir Ernest Barker, at the age of eighty-five, means not only the loss of a great scholar and teacher but also the loss of a symbol of what may well seem, in 1960, to have been a happier and more confident world. Ernest Barker always denied being a 'scholar' in the old-fashioned Oxford and Cambridge classical sense. But all that that meant was that he was not one of the elaborately skilful composers in the ancient tongues whom Macaulay (who himself was a master of these dexterities) made fun of. Barker was a very distinguished product of the Oxford 'Greats' school. I suspect that he never lost his conviction, even after many happy years in Cambridge, that for the statesman and for the teacher of statesmen the school of *Literae Humaniores* was the ideal education.

It would be both absurd and unjust to suggest that Barker was an old-fashioned classic incapable of understanding or appreciating the contributions of more modern studies to the science (or as he would have preferred, I think, to put it) the art of politics. He preserved into his old age a great deal of curiosity about modern developments and modern contributions. But it would be also absurd to pretend that he was ever convinced that the modern American statistical analysts of political behaviour and, still more, the modern psychological explainers of political behaviour, have really improved on the study and explanation of the human situation provided by Plato, Aristotle, and St. Thomas Aquinas.

Barker wrote a great many books, all of them meritorious and some very remarkable. His essay on "The Crusades" for the "Encyclopædia Britannica", published as a little book, seems as remarkable to me now as it seemed when, as an undergraduate in Oxford, I had my attention directed to it by the late B. H. Sumner. In his old age he wrote what I am told was an extremely competent book on Byzantium. But I think that his greatest achievement was his "Political Thought of Plato and Aristotle"—a book full of learning and wisdom. I once reproached him for not producing a new edition and almost coaxed him into an admission that he had wasted a good deal of time on less-important things. He did, in fact, produce a new volume on Plato, but never finished the complete revision of this masterpiece. He spent (or, as I think, wasted) an immense amount of learning on a translation of Gierke, which involved the resuscitation of a great many rightfully forgotten Germans. 'C'était magnifique', but it was not what we wanted from Ernest Barker. I think he came to feel this, for he does not cite this laborious and learned and in some ways useful work in "Who's Who".

Barker was a Liberal (both upper case and lower case letters), and that comes out in the admirable little volume he wrote in the Home University Library on English political thought "From Herbert Spencer to To-day" and in his "Principles of Social and Political Theory". He would never have understood that it was possible to separate politics from morals, to reduce politics to statistical description. As much as Dr. Gunnar Myrdal, he would have held that the idea of ethically neutral means was nonsense. Politics was about ethics. If this were not true, it was not worth serious study.

Barker was not only a man of prodigious learning, he was also a man of prodigious energy. At an age when most people think of retirement as a reality, nearly ten years after he had ceased to be professor of political science in Cambridge, he went to Germany, out of a strong sense of mission, to rehabilitate the lost German tradition of academic freedom. It was a task that might have daunted a younger man, and he suffered a very serious illness from which, some think, he never quite recovered, although he continued to work with an energy that would have shamed many men much younger than he was.

In addition to immense intellectual and physical energy, Barker was extremely handsome and had a most magnificent voice, which was made even more effective as it preserved its northern burr. There is a tradition in Peterhouse that he imagined he had lost this burr. Fortunately there was no basis for this illusion. He not only had an academic career of great distinction, but also one of great variety. From Manchester Grammar School he went to Balliol and was successively Fellow of Merton, St. John's, and New College, where he had a most admiring pupil in the late Harold Laski. He was, for a short and not very happy time, principal of King's College, London, and then he was, for a very long and happy time, the first professor of political science in Cambridge and Fellow of Peterhouse. In contemplating Ernest Barker it was easy to think of him as a relic of a dead past. But it was much wiser to think of him as a representative of a great past, of the disciples of T. H. Green (and of Henry Sedgwick), of the moralistic and learned political philosophers of a great era of modern English history. He survived into what he must have thought—and did think—was an evil and distracted world. But he continued to be loyal to both his theory and practice. If sometimes he seemed unreceptive to new ideas, he was totally above the temptations of fashion or current academic prestige. "We shall not look upon his like again."

D. W. BROGAN

Dr. Hsien Wu

DR. HSIEN WU, an authority on blood analysis, died on August 8, 1959. He was born in Foochow, China, on November 24, 1893. He attended a local high school in Foochow and then enrolled in the naval architecture course at the Massachusetts Institute of Technology. After two years, he changed his course to chemistry and was awarded the B.S. degree by the Institute in 1916. After remaining as an assistant in the Department of Chemistry for a year at the Institute, he entered Harvard Medical School in 1917 for graduate study with Dr. Otto Folin in the Department of Biological Chemistry, and received the Ph.D. degree in June, 1919. He continued his researches as a Fellow with Dr. Folin for a further year.

In 1920, Dr. Wu returned to China as assistant in biochemistry at Peiping Union Medical College. After one year, he was raised to an associate and later placed in charge of the Department. In 1924, he was appointed an associate professor of biochemistry and in 1928 he was raised to professor and chairman of the Department of Biochemistry. He continued as chairman of the Department until January, 1942, when Japan took over the College. In March, 1944, he was called to Chungking to organize the Division of Nutrition and join the National Institute of Health. In July, 1944, the Chinese Government sent him to the United States as its nutrition expert on a special mission to study rehabilitation reconstruction programmes for post-war China. He served as director of the Nutrition Institute, Ministry of Health, until the end of 1947. He was also associated with the Institute as the director of its Peiping Branch during 1946–47.

In 1947, Dr. Wu and five other Chinese scientists were invited by Unesco to attend the International Physiological Congress which was held at Oxford. On his return from the Congress, by way of the United States, in September, 1947, he was extended the privileges of a visiting scholar in the Department of Biochemistry, College of Physicians and Surgeons, Columbia University. In June, 1948, he was appointed a research associate at Columbia. Sensing the political situation in China, he finally made preparations to move his family to the United States.

Dr. Wu joined the Department of Biochemistry, Medical College of Alabama, Birmingham, on September 1, 1949, as visiting professor. Following a heart attack in October, 1952, he retired on September 30 and moved to Boston, Massachusetts, where he devoted his time to higher mathematics, Spanish and writing.

Dr. Wu was a member of an Administrative Committee of three functioning as director of Peking Union Medical College, 1935–37. He was a member of the Chemistry Division of the National Committee on the Standardization of Scientific Terminology during 1921–27; he served as editor of the *Chinese Journal of Physiology*, 1926, and continued on the editorial board until 1942; and was president of the Chinese Physiological Society, 1931–34.

Dr. Wu was a tireless investigator. His bibliography has 160 entries, which includes two books: "Principles of Nutrition" (1929) and "Principles of Physical Biochemistry" (1934). His fields of research included blood chemistry, gas and electrolyte equilibria, nutrition, immunochemistry, proteins and the metabolism of amino-acids using amino-acids labelled with nitrogen-15 as tracer.

In addition to his contribution of the famous system of blood analysis he also was a pioneer in his researches and theory in other major areas. He made quantitative analyses of the precipitate from the antigen–antibody reaction following the use of labelled antigen. In 1928, he used hæmoglobin which was labelled with a coloured group. The next year, he reported the use of iodo-albumin as the antigen and he analysed the antigen precipitin precipitate for iodine and nitrogen content. These were the first analyses of this nature and antedated the use of isotopes for such analyses by several years. He also proposed the first reasonable theory of denaturation of proteins in 1931. Other valuable contributions to protein chemistry by Dr. Wu were his studies on the molecular weights and osmotic pressures of hæmoglobins of experimental animals in aqueous solution, 65 per cent glycerine solution and in concentrated urea. He noted that the molecular weights of oxyhæmoglobins and hæmoglobins of dog and sheep are not changed in urea solution whereas those of ox and horse are reduced to one-half.

EMMETT B. CARMICHAEL

NEWS and VIEWS

Pioneer 5

THE United States space probe *Pioneer* 5 was launched from Cape Canaveral in Florida at 13.00 U.T. on March 11, to become the third artificial planet of the Sun. The launching missile was a three-stage *Thor-Able* rocket, and the final-stage vehicle attained a velocity of about 6·9 miles per sec. relative to the Earth, in a direction nearly opposite to that of the Earth's motion around the Sun. Its residual velocity relative to the Sun, after escaping from the Earth's sphere of influence, was therefore less than the Earth's orbital speed, and its orbit is consequently smaller than that of the Earth: it is expected to be about 75 million miles from the Sun at perihelion, and about 92 million miles at aphelion, with a period of 311 days. At perihelion it will be near the orbit of Venus, which moves in a nearly circular path at a distance of 67 million miles from the Sun.

Pioneer 5, which weighs 90 lb., has a 5-watt radio transmitter for use near the Earth and a 150-watt transmitter for use at greater distances. The power is provided by solar batteries arranged in four 'paddle-wheel' vanes. To conserve energy, the transmitter is to be switched on for only a few minutes each day, in response to commands from Earth. The radiotelescope at Jodrell Bank is the main tracking station, and the 150-watt signals should be received at distances out to about 50 million miles. If so, *Pioneer* 5 will be within communication distance for about the next five months. *Pioneer* 5 carries various scientific instruments, including a high-energy radiation counter, an ionization chamber and Geiger counters to measure medium-energy radiation, a magnetometer to determine both the strength and direction of the magnetic field in space, and equipment for detecting micro-meteorites.

Pharmacology at University College, London: Prof. F. R. Winton

PROF. F. R. WINTON, who is retiring from the University of London chair of pharmacology at University College, was trained originally in chemistry and had been working for some time as an industrial chemist with the Mond Nickel Co. when he decided to take up medicine. He graduated M.D. (Cambridge) in 1926, and he obtained the D.Sc. (London) in 1942. After graduation, he worked with A. J. Clark in the Pharmacology Department of University College, London, and in 1931 returned to the Department of Physiology, Cambridge, first as lecturer and then as reader. In 1939 he was appointed to the London chair of pharmacology at University College, in succession to Prof. J. H. Gaddum. His earliest scientific work, carried out in conjunction with L. T. Hogben, dealt with the identification of a frog melanophore-expanding hormone in the pituitary gland. His subsequent scientific work was concerned mainly with two fields, the physiology and pharmacology of the kidney and of smooth muscle. His chief interest lay in the physical factors controlling the activities of the kidney and he succeeded in establishing and supporting experimentally a series of remarkably simple relations between arterial, venous, ureteric and glomerular pressures from which the capillary glomerular pressure of the mammalian kidney could be calculated. He also worked on the mode of action of diuretics, and during the War he was engaged in a study of the renal damage which followed crush injury in the limbs. Much of his renal work is summarized in a Harvey Lecture given in 1951. Winton's studies on smooth muscle reveal another aspect of his intense interest in the physical aspects of biological processes. They represent one of the earliest systematic studies of 'tonus' and contractility in invertebrate and smooth muscle. During the War, Winton was the only head of a department left in the Medical Faculty of University College and acted as dean throughout. The success of the evacuation of the Faculty to Leatherhead was in great measure due to his personality and drive. He has published several books, among which is the widely used "Text-book of Physiology", by Winton and Bayliss. Winton is a man of varied interests, a connoisseur of wine and a 'cellist of well above average amateur status. He has been throughout, together with his wife, the violinist, Bessie Rawlins, the main inspirer and one of the principal performers of the Chamber Music Club at University College. He has taken a major part in establishing the Student Health Service in the College.

Prof. H. O. Schild

H. O. Schild, who will succeed Prof. Winton, was born in Fiume. He studied medicine in Munich and began his career in pharmacology in Straub's laboratory. In 1932 he went to London to work with Sir Henry Dale at the National Institute for Medical Research. Later, he worked at the University of Edinburgh with I. de Burgh Daly and the late A. J. Clark. In 1937 he went to University College, London, to join Gaddum, with whom he had worked when he first came to England. He was appointed reader in 1947. Working with Gaddum, Schild found that adrenaline in high dilution, in contrast to noradrenaline, gave a strong green fluorescence in alkaline solution. This reaction, modified in various ways, is still the most sensitive physical method for the assay of adrenaline. Schild was the first to infer the presence in extracts of the suprarenal medulla of a second pressor substance, which we now know to be noradrenaline. He developed a method of demonstrating release of histamine in anaphylaxis by estimating the histamine diffusing out of small tissue particles suspended in Ringer solution on the addition of antigen. With this simple and ingenious method he showed with Hawkins and others for the first time that histamine is also released from lungs of asthmatic patients by the specific allergens. In collaboration with Mongar, he became interested in the intracellular reactions with lead to release of histamine. Schild observed that the anaphylactic and allergic contractions of smooth muscles cannot be fully accounted for by the release of histamine, since they occurred also in muscles rendered insensitive to it. We also owe to him the knowledge that strychnine and strychnine methiodide have the property of releasing histamine, one of the earliest demonstrations of histamine liberators; also the finding that inhibitors of diamine oxidase potentiate histamine just as anticholinesterases potentiate acetylcholine.

Pharmacologists value most perhaps his contributions to bioassay. He introduced statistically controlled methods for the assay of substances on isolated tissue preparations, whereby it became possible not only to obtain more accurate results but also to estimate the limit of error in each assay. They are used, for example, for the B.P. assay of oxytocin. He introduced a method for quantitative measurement of the activity of oxytocic drugs on the post-partum uterus in women. In order to ensure greater accuracy when assaying substances on smooth muscles, he designed an automatic method which is widely used. His approach to the study of drug antagonism has been particularly fruitful. It is the *pAx* method by which drug antagonism can be expressed quantitatively and agonists classified by means of antagonists. Drugs which act on the same receptors give the same *pAx* value with antagonists. Recently, he made the intriguing observation that drugs contract the fully depolarized smooth muscle, thereby excluding membrane depolarization as the only cause of contraction.

Meteorological Department of India:

Shri S. Basu

Shri Basu retired from the Indian Meteorological Service on February 23 after thirty-two years of service in that Department. He was born in the United Provinces on October 23, 1903, where he spent his early years and had a brilliant educational career. He completed his university career at Allahabad under the guidance of the late Prof. Meghnad Saha, whose personality profoundly influenced him as a young student. After completing his post-graduation in physics, Shri Basu entered the Meteorological Department of India in 1927, where he served until his retirement last month. During the period he was director-general of observatories, the Meteorological Department made notable advances in many directions. Apart from his activities in meteorological spheres, he actively participated in some of the other scientific organizations in India; he is a Fellow of the National Institute of Sciences in India and has been the secretary of it during the past four years. He is also a Fellow of the National Academy of Sciences, Indian Physical Society and a member of the Aeronautical Society. He is also the president of the Association of Indian Geographers. He was the founder-president of the Indian Meteorological Society and is now one of the vice-presidents.

In 1951 he was elected vice-president of the Commission for Climatology of the World Meteorological Organization. As representative of India, he led the Indian delegation at the meeting of the World Meteorological Organization in 1955 and 1959 at Geneva. He was a member of the Executive Committee of the World Meteorological Organization during 1954–59. As president of the Regional Association II of the World Meteorological Organization for Asia (1954–59), Shri Basu did a great deal of preliminary work the influence of which will serve to guide the affairs of the Association for many years to come. He has also been India's member in the International Civil Aviation Organization and of the Special Committee of the International Union of Geodesy and Geophysics for the International Geophysical Year.

Shri P. R. Krishna Rao

SHRI P. R. KRISHNA RAO succeeds Shri S. Basu as the director general of observatories, Government of India. He has had long experience of work in the department, having entered the service thirty-three years ago. He has had experience of international meteorological conferences since 1946, when he attended the first International Meteorological Organization Conference of Directors held in London after the Second World War. He recently led the Indian delegation to the joint session of the Meteorological Division of the International Civil Aviation Organization and the Commission for Aeronautical Meteorology of the World Meteorological Organization held in Montreal, and later paid short visits to the Canadian and United States Meteorological Services at Toronto and Washington.

Botany at Belfast : Prof. D. J. Carr

DR. D. J. CARR has been appointed professor of botany in the Queen's University of Belfast in succession to Prof. J. Heslop Harrison, who has been appointed professor of botany in the University of Birmingham (*Nature*, 173, 150; 1954). Prof. Carr was educated at the University of Manchester after war service with the Royal Air Force. In 1949 he became an assistant lecturer at Manchester, and after a year at the Max Planck Institute at Tübingen, working in association with Dr. G. Melchers, he was appointed a senior lecturer in the University of Melbourne. His promotion to a readership at Melbourne followed in 1959. While at Manchester and Tübingen, Carr was engaged in work on developmental physiology, especially in relation to the photoperiodic responses of flowering plants. Among his contributions in the field of photoperiodism was a demonstration, using grafting techniques, that leaves cannot be photoperiodically induced unless they are in organic connexion with a shoot apex, a result interpreted as favouring Gregory's hypothesis of photoperiodic induction, according to which there can be no accumulation of a 'flowering hormone' in leaf tissues. In Australia, Carr's interests have ranged widely, penetrating into the fields of ecology and taxonomy. He has added to the Australian bryophyte flora a remarkable member of the Marchantiales, *Monocarpus sphaerocarpus* D. J. Carr, a plant of saline flats in north-western Victoria. In association with his wife, Dr. S. G. M. Carr, he has recently begun a major study of the developmental morphology of the inflorescence and flowers of the important genus *Eucalyptus*.

Commonwealth Assistance in Training Teachers

IN a written answer in the House of Commons on February 24, the Secretary of State for Commonwealth Relations, Mr. C. J. M. Alport, said that good progress was being made in carrying out the proposals for assistance in training teachers put forward by the United Kingdom at the Commonwealth Education Conference. By the beginning of the next academic year it was expected that more than four hundred additional Commonwealth students would be coming to the United Kingdom to take courses in teacher training institutions with bursaries provided by the United Kingdom Government covering their tuition fees and full maintenance. This was more than half the existing total of Commonwealth teachers already receiving training in Britain. Places would be available for these students in a wide variety of training courses at institutions throughout the United Kingdom, and arrangements for their reception and welfare would be made by the British Council.

Mathematics and Science Teachers in Great Britain

IN a written reply to a question in the House of Commons on February 25 regarding mathematics and science teachers, Sir David Eccles said that in 1959 there were 5,100 mathematics and 7,700 science graduates teaching senior pupils in maintained schools, as well as 3,250 non-graduate specialists in these subjects in schools with courses leading to the General Certificate of Education at Ordinary level. Inquiry in 1958 revealed 244 vacancies in these schools for mathematics teachers and 261 for science teachers. Assuming an annual net increase of 500 mathematics and science graduates, the shortage of these teachers on 1959 staffing standards would be 500 in 1961, 750 in 1962, 900 in 1963 and 350 in 1964. In 1965, 250 would be available to improve on 1959 standards. The actual shortage would depend on the results of present measures to increase the number of graduates and the output of specialists from training colleges.

Laboratory Accommodation in British Schools

IN reply to a question in the House of Commons on February 11, the Minister of Education, Sir David Eccles, said that about £5 million would be spent in the 1960–61 and 1961–62 major building programmes to improve laboratory accommodation in existing schools; in addition, new schools had the full range of science facilities. He said he regarded this provision as one of the most important elements in the building programme, and in the maintained schools we were building, and would continue to build, every year at a higher rate than ever was spent on the public schools.

The Library Service in Great Britain

REPLYING in the House of Commons on February 11 to a question regarding the introduction of legislation to implement the recommendations of the Roberts Committee on the Public Library Service, the Minister of Education said that the Government accepted the need for legislation in this field, and he hoped shortly to begin discussions with associations of local authorities on his proposals. He thought it wise to obtain the views of local authorities before the subject was discussed in the House, and agreed that it was necessary to take into account the view that no good local authority that is running a virile library service should be deprived by its size of the right of continuing to do so. Sir David Eccles undertook to bring into his discussions the Smaller Library Group of the Library Association. He indicated that his proposals were more in the sense of retaining the power, authority and responsibility of smaller local authorities, and not transferring all these responsibilities to county councils, and that the library service essentially should be decided by local authorities and not subject to national standards other than the proposals of the report. He also agreed that account must be taken of the possibility of changes in the structure of local government resulting from the recommendations of the Commissions now operating before making any alteration in the library service and that the problem was highly controversial.

Fortieth Anniversary of the Liebig Museum

The Liebig Museum, Geissen, celebrates this year its fortieth anniversary. Although the Museum was not inaugurated until 1920, the idea had long been in the minds of German scientists and, but for the intervention of the First World War, would have most probably materialized many years before. It was decided at the beginning of the present century to set up as a memorial the Laboratory which Justus Liebig had made so famous. In 1909 a committee was formed for this purpose, including such scientists as Prof. Volhard, who at the time was the last living assistant of Liebig, Prof. Sommer, Prof. Behagel, Dr. Merck and many others. It was from this committee that the Liebig Association emerged in 1911. Much of Liebig's original equipment was transferred from the new Institute to the original Laboratory which he had persuaded the Darmstadt government to provide at Geissen in the early nineteenth century. Repair work was carried out on the Laboratory and much of the lost equipment was copied on the strength of drawings which had been found. The First World War delayed the opening until March 26, 1920; but such was its fate that only twenty years later it suffered further damage by yet another World War. However, after thorough reconstruction, for which funds could only be raised by collections, the Museum was reopened during the summer of 1952; the Institute which had proved to be the mother of chemical institutes all over the world was once more established.

U.S. Federal Funds for Science

The eighth issue by the National Science Foundation of Federal Funds for Science, dealing with the Federal Research and Development budget for 1958, 1959 and 1960, estimates Federal expenditure on scientific research and development for 1960 at 8,100 million dollars, an increase of 3 per cent over 1959, and compared with a 34 per cent increase in 1959 (pp. iii+74. NSF-59-40. Washington, D.C.: Government Printing Office, 1959. 65 cents). In 1959 the Department of Defence, the Atomic Energy Commission and the National Aeronautics and Space Administration accounted for 6,700 million dollars out of the 7,200 million for research and development. Of the remainder, 244 million dollars was expended by the Department of Health, Education and Welfare, 119 million dollars by the Department of Agriculture, 62 million dollars by the Department of the Interior and 57 million dollars by the National Science Foundation. Profit organizations were responsible for 65 per cent of Federal expenditure on research and development, Federal organizations for 24 per cent, and educational institutions for 9 per cent. Of the estimated expenditure of 1,444 million dollars on research in 1959, 964 million dollars was for the physical sciences, 432 million dollars for the biological, medical and agricultural sciences, and 48 million dollars for the social sciences. Fundamental research accounted for an estimated 488 million dollars in 1959 compared with an actual 331 million dollars in 1958 and an estimated 494 million dollars in 1960; of this, 335 million dollars were for the physical sciences, 142 million dollars for the biological sciences, and 11 million dollars for the social sciences; 49 per cent of the total was expended through educational institutions, 5 per cent through profit organizations and 37 per cent through Federal organizations. Expenditure on research and development plant increased from an actual 342 million dollars in 1958 to an estimated 662 million dollars in 1959 and 447 million dollars in 1960. Of the latter figure, 136 million dollars in 1959 and 92 million dollars in 1962 were on non-Federal sites, and 531 and 355 million dollars, respectively, on Federal sites. Since 1951, basic research, as a proportion of total research, has increased from about one-fourth to almost one-third. In the same period, expenditure on the biological sciences increased by 118 per cent, on the physical sciences by 56 per cent, and on the social sciences by 60 per cent.

Funds for Research in the United States

Data now available from the U.S. National Science Foundation's continuing survey of funds for research and development in the United States indicate an increase in those funds from just over 5,000 million dollars in 1953–54 to 12,000 million dollars in 1959–60, due primarily to an increase of about 160 per cent in the funds used for research and development by private firms and certain types of related organization which constitute the "industry sector" of the Foundation's programme: for industry the rise is expected to be 3,600 million dollars to 9,400 million dollars, more than half of which now comes from the Federal Government. For colleges and universities the corresponding increase is 126 per cent (450 million dollars to 1,000 million dollars), and for other non-profit institutions, 140 per cent (100 million dollars to 250 million dollars). Taking the completed year 1957–58 in comparison with 1953–54, funds for research and development increased from 5,150 million dollars to 10,050 million dollars, and of these Federal Government agencies provided 6,130 million dollars, compared with 2,740 million dollars; for industry the corresponding figures are 3,660 million dollars and 2,240 million dollars; for colleges and universities, 180 million dollars and 130 million dollars; and for other non-profit institutions, 80 million dollars and 40 million dollars. In terms of performance it is estimated that industry was responsible for 76 per cent of research and development in 1956–57 compared with 70 per cent in 1953–54; Federal Government agencies for 15 per cent, compared with 19 per cent; colleges and universities for 7 per cent, compared with 9 per cent; and other non-profit institutions were still responsible for 2 per cent.

The Metallurgist

By the issue of the first number of *The Metallurgist*, the Institution of Metallurgists has at last a publication worthy of its growing size and importance (*The Metallurgist*, 1, No. 1; January 1960. Incorporating the *Bulletin of the Institution of Metallurgists*. Pp. 64. Annual subscription, 63*s*. post paid). It is no function of the Institution to duplicate the journals of the other metallurgical institutes, and the contents of this issue are, therefore, concerned either with matters of domestic importance or with articles of general appeal. Among the former may be included the Institution's revised examination requirements, a most carefully carried out piece of work, and reports on the results of the examinations in 1959 and on national certificate results for which it is responsible. The articles comprise Prof. A. J. Murphy's presidential address, "Some Recollections" by Dr. Harold Moore, the first president, and a most interesting article by Prof. R. W. K. Honeycombe entitled "New Horizons in

Metallography", which discusses the applications of the electron microscope and of electron-probe micro-analysis. Dr. Howard K. Warner contributes an account of metallurgy in Australia which will be of great value to any member who contemplates taking up an appointment, teaching or industrial, in that continent. The presentation of the journal is most attractive and the illustrations excellently reproduced, the whole being a credit both to the Institution itself and all those responsible for its appearance.

Science in the School Library

A NEW edition of "Science Books for a School Library" has been published by John Murray for the Science Masters' Association (1959. 3*s.* 6*d.* net). It contains in its 54 pages three large sections on biology, chemistry and physics, of about 150, 100 and 100 titles, respectively, and smaller lists, on the history of science, astronomy, geology and meteorology. The final section is a list of books suitable for a secondary modern school science library. About half the entries have been reviewed in *The School Science Review*, and the number of the issue is given. Annotations are attempted in a rather half-hearted way, and the need for brevity is no excuse for "a readable book", "a well written history", "a most interesting book", or "packed with information". One wonders why J. B. Conant's "On Understanding Science" is in the chemistry section and hopes that all schools are able to afford Singer's "A History of Technology" (5 vols., £42). The list will be of great use to new schools setting up a library for the first time, or as a check list for an established library. Both users and the editors of the next edition would find of great interest two similar works—"The A.A.A.S. Science Book List" (American Association for the Advancement of Science, 1959) and the National Book League's "Science for All".

Tunnel Diodes

IN a recent press release, Standard Telephones and Cables, Ltd., announced the development of a new semiconductor device known as a tunnel diode, believed to be the first of its kind produced in Europe. The main feature of tunnel diodes is that they exhibit a marked negative resistance characteristic, which is likely to have important applications in radio circuit technique. The fact that a germanium diode made of very highly doped material can exhibit a negative resistance for a small applied voltage (100–200 mV.) was first reported by L. Esaki (*Phys. Rev*, 109, 603; 1958). The results of a theoretical and experimental examination of tunnel diodes as devices for use at very high frequencies were described by H. S. Sommers, of the R.C.A. Research Laboratories, Princeton, U.S.A. (*Proc. Inst. Radio Eng.*, 47, 1201; 1959). Diodes with a negative conductance of 1 mho have been made and these oscillate at frequencies above 1 kMc./sec. when incorporated in a special microstrip mounting of low series impedance. Apart from its use as a self-excited oscillator, the tunnel diode, when mounted in a coaxial or microstrip line, can be used as a switch with an operating period of a few milli-micro-seconds. It is believed that a different approach to electronic circuit problems is necessary in order to exploit fully the application of these tunnel diodes; and it is with the object of assisting circuit design engineers to gain experience of these that samples suitable for use at frequencies up to about 5 Mc./s. are being made available in Great Britain by Standard Telephones and Cables, Ltd. Development work is proceeding rapidly, however, and it is expected that devices with a more advanced performance will shortly become available.

High-atmosphere Research in the Meteorological Office

THE importance of the continuous study of conditions in the highest parts of the undissociated atmosphere (between about 30 and 100 km.) is now recognized among meteorologists. To further such studies, the Meteorological Office has set up a new assistant-directorate, with responsibilities for both theoretical and experimental research. It is intended to investigate the composition and physical state of the atmosphere up to about 100 km. with special balloons and, it is hoped, small rockets. In addition, the new unit will take over the present design studies for meteorological observations from the second *Scout* satellite. Dr. R. Frith, formerly assistant-director in charge of research in atmospheric physics, has been placed in charge of the unit.

James Clayton Prize, 1959

THE James Clayton Prize for 1959 has been awarded to C. Bernard Dicksee, for his pioneering and subsequent development and research work on the high-speed compression-ignition engine, and Prof. G. Fernie Mucklow, for continuous research and experimental work into the propagation of pulses (wave action) in gases, and problems connected with the internal combustion engine. This Prize is awarded annually to a member or members of the Institution of Mechanical Engineers, who contribute most in the year to modern engineering science by way of research, invention, experimental work or a treatise or paper on a modern engineering subject, or originality in engineering design, or by service to engineering.

The Corday-Morgan Medal and Prize

PROF. C. KEMBALL, professor of physical and inorganic chemistry, the Queen's University of Belfast, has been awarded the Corday-Morgan Medal and Prize for 1958 of the Chemical Society, in consideration of his contributions to the study of heterogeneous catalytic reactions. The award, consisting of a Silver Medal and a prize of 200 guineas, is made annually to the chemist of either sex and of British nationality who has published during the year in question the most meritorious contribution to experimental chemistry, and who has not, at the date of publication, attained the age of thirty-six years. Applications in respect of the award for 1959 must be received by the General Secretary of the Chemical Society, Burlington House, Piccadilly, London, W.1, not later than December 31, 1960, and for the award for 1960 before the end of 1961.

Nature Conservation: Postgraduate Diploma Course

THE social, economic and political problems arising from the results of a shortsighted policy for land use have been painfully obvious for a long time. Practical object lessons on a vast scale can still be seen in many parts of the world. Conservation involves the long-term study of land use, and one of the main problems is the prediction of the immediate and ultimate effects of any programme of agriculture or resource exploitation. The solution to these problems will provide the means for avoiding the disasters of the past. One of the main difficulties has been the lack of trained men, and to meet this need a postgraduate diploma course in conservation will be inaugurated at University College, London, in July. The course will consist of an initial period of field work, extensive and exploratory in nature, followed by two terms of indoor teaching and finishing with a second period of more intensive field work. The course will be run by the Botany Department, in collaboration with Geography and Zoology Departments of University College, with assistance from other departments and a wide range of seminars. The syllabus will include the physical factors of the environment, plant and animal ecology, and a study of different forms of land use and their effect on soils, plants and animals. This fundamental grounding should be applicable to conservation problems in different parts of the world. The Nature Conservancy will award maintenance grants for students to attend the course and will assist in other ways, especially with field work. The diploma will be a useful qualification to those seeking jobs with the Nature Conservancy, or similar organizations in other countries, or with the Colonial Service, or in any other aspect of applied ecology or land use.

Summer School in Radiation Protection

A fortnight's summer school of health physics (radiation protection) will be held at the Imperial College of Science and Technology, London, starting on July 4. The course will offer training in radiation protection, at postgraduate-level, for persons who are directly concerned with the subject, and should be of interest to safety officers, medical officers of health, factory inspectors and others concerned with radiation protection. The emphasis will be on practical applications, and training in the measurement of radioactivity will be included. Further information can be obtained from the Registrar, Imperial College of Science and Technology, South Kensington, London, S.W.7.

Royal Society of Tasmania

The following officers of the Royal Society of Tasmania for the year 1960 have been announced: *President*, His Excellency the Governor of Tasmania, the Right Honourable Lord Rowallan; *Vice-Presidents*, Dr. D. Martin and Mr. R .M. H. Garvie; *Council Members*, Mr. D. H. Borchardt, Dr. C. Craig, Mr. W. F. Ellis, Dr. E. R. Guiler, Mr. L. W. Miller, Mr. K. D. Nicolls, Mr. M. S. R. Sharland and Mr. J. W. C. Wyett; *Hon. Secretary and Librarian*, Dr. W. Bryden; *Hon. Treasurer*, Mr. G. E. A. Hale; *Hon. Auditor*, Mr. A. M. D. Hewer.

The Night Sky in April

Full moon occurs on April 11d. 20h. 28m. U.T., and new moon on April 25d. 21h. 45m. The following conjunctions with the Moon take place: April 1d. 08h., Aldebaran 0·4° S.; April 16d. 19h., Jupiter 5° S.; April 17d. 21h., Saturn 4° S.; April 22d. 01h., Mars 2° S.; April 24d. 01h., Mercury 1° S.; April 28d. 16h., Aldebaran 0·5° S. Mercury, Venus and Mars are too close to the Sun for easy observation. Jupiter is a morning star, rising at 1h. 40m., 0h. 50m. and 23h. 45m. on April 1, 15 and 30, respectively. It is moving eastwards in Sagittarius until April 20, on which date it is stationary; afterwards it begins to move westwards among the stars. Its stellar magnitude is − 1·9 and its distance from the Earth in the middle of the month is 450 million miles. Saturn is also a morning star, moving eastwards in Sagittarius until April 27, when it is stationary and commences to move westwards among the stars. It rises at 2h. 35m., 1h. 40m. and 0h. 45m. on April 1, 15 and 30, respectively. Its stellar magnitude is + 0·7 and its distance from the Earth on April 15 is 910 million miles. Occultations of stars brighter than magnitude 6 are as follows, observations being made at Greenwich: April 6d. 01h. 00·8m., 29 Cnc. (*D*); April 28d. 15h. 44·0m., Aldebaran (*D*); April 28d. 17h. 03·0m., Aldebaran (*R*); *D* and *R* refer to disappearance and reappearance, respectively. The Lyrid meteors are active during April 20–22; the radiant is at R.A. 18h. 04m., Dec. + 33°, and conditions for observation are fairly favourable.

Announcements

Mr. S. S. Kind has been appointed editor of the newly founded *Journal of the Forensic Science Society*. Communications should be addressed to the Editorial Office, c/o Rossett Holt, Pannal Ash Road, Harrogate, Yorks.

The Bureau of American Ethnology of the Smithsonian Institution has announced recently (Bulletin No. 130) completion of the long awaited index to the volumes entitled "Handbook of South American Indians". The index appears in Vol. 7 (Washington, D.C.: Government Printing Office, 1959).

Following an investigation into technical readership by the Department of Scientific and Industrial Research, Heywood and Co., Ltd., are to establish *Electronics Weekly* in September. It will appear as a newspaper and will be directed at manufacturer and user, as well as to management and scientific staff. Further information can be obtained from Heywood and Co., Ltd., Drury House, Russell Street, London, W.C.2.

A symposium on the Matrix will be held by the Bone and Tooth Society at the Nuffield Department of Orthopædic Surgery, University of Oxford, on March 29. Further information can be obtained from B. Cohen, Royal College of Surgeons, London, W.C.2.

The spring meeting of the Parasitology Group of the Institute of Biology is to be held at the Liverpool School of Tropical Medicine, Pembroke Place, Liverpool 3, during April 6–8. Further information can be obtained from L. P. Joyner, Ministry of Agriculture, Fisheries and Food, Central Veterinary Laboratory, New Haw, Weybridge, Surrey.

An informal discussion by the Colloid and Biophysics Committee of the Faraday Society on Cytoplasmic Particles and their Role in Protein Synthesis will be held in the University of Reading during March 28–29. Further information can be obtained from Prof. J. A. V. Butler, Chester Beatty Research Institute, Fulham Road, London, S.W.3.

The Campaign for Nuclear Disarmament announces the formation of a Scientists Group, the purpose of which is to advise the Campaign, in the light of available scientific information, on such subjects as fall-out, civil defence, control of tests, effects of nuclear war, relevant aspects of general disarmament, and civilian uses of nuclear energy. The Group is publishing a regular bulletin, of which the first number (4 pp.) has recently appeared; it includes a number of abstracts of recent scientific papers and publications dealing with fall-out and related topics. Further information can be obtained from the Secretary, Campaign for Nuclear Disarmament, 2 Carthusian Street, London, E.C.1.

UNIVERSITY EDUCATION IN CENTRAL AFRICA

THE events which led up to the granting of a Royal Charter to the University College of Rhodesia and Nyasaland on February 11, 1955, have been reviewed in *Nature* (172, 1; 1953. 173, 107; 1954. 175, 181; 1955). Now, five years after its establishment, the first results of degree examinations in the College have been announced, and it is opportune to review the five years of life of this flourishing multi-racial establishment. A basic principle in the foundation and planning of the University College was that there should be no discrimination in it on racial grounds, and, as was said in 1955 (*Nature*, 175, 181; 1955), "In a country where racial tension can arise, the position of a multi-racial University is especially important". The agreement of Her Majesty Queen Elizabeth the Queen Mother to become the first President of the College, and her presence at Salisbury when she was formally installed on July 5, 1957, were particularly happy auguries for the development of this young university institution. As President of the College she will preside at the degree ceremony on May 13, 1960, in Salisbury.

The first academic session opened in March 1957 with a total of 74 students, 8 of whom were African (*Nature*, 179, 612; 1957). During the session that has just closed there were 168 students, of whom 38 were African. The proportion of African students in the College is steadily rising, and, with the development of sixth-form school education throughout the Federation of Rhodesia and Nyasaland, may be expected to rise still further.

The University College is in special relationship with the University of London and the students therefore now take the degree examinations of that University. At the end of the session which has just closed 19 candidates sat for the B.A. General degree, of whom 17 were successful, one being placed in the first division. Six candidates out of the seven who sat for the B.Sc. General examination, and both of two candidates who took that for B.Sc. Agriculture, were successful in obtaining degrees. In addition, 23 out of 26 candidates successfully completed the course and examination for the postgraduate certificate in education. Of the total number of successful candidates, nine were African.

The examination results to date are a useful index of the soundness of the academic progress of the College. Nevertheless, the needed growth in breadth and depth of educational facilities still depends upon financial encouragement, and it is with great satisfaction that the news has recently been received of an offer of a grant of £250,000 by the Nuffield Foundation towards the establishment of a Medical School at the College. Training in medicine is extremely expensive, and it was natural that the institution of a Medical School should be delayed until a number of other faculties had been established, although this ultimate development was contemplated from the start.

The generous offer from the Nuffield Foundation is made on the understanding that the teaching hospital for the Medical School takes the form of a new multi-racial hospital on the site of the University College. The University of Birmingham and the University College of Rhodesia and Nyasaland have agreed that when a Medical School is established at Salisbury it shall initially be an affiliated institution of the University of Birmingham. This means that, for the period of affiliation, the students of the Medical School of the University College will be prepared for the degrees in medicine of the University of Birmingham just as those in the Faculties of Arts and Science at present prepare for the degrees of the University of London.

As so often seems to hold for university institutions, the financial support of the University College is scarcely adequate for all that, on other grounds, it might hope to do. Her Majesty's Government, which in the past has provided capital money totalling £1,475,000, has recently made a further generous grant of £1,400,000 towards capital costs for the College. The Government of the Federation of Rhodesia and Nyasaland has provided £750,000 over the first five years, for recurrent expenditure, and one may hope that it will give further generous support during the new quinquennium to help to provide for the cost of the much needed expansion which is already planned. At present, besides medicine, the College lacks only engineering and theology among the major faculties to be expected in a university institution.

The working out of a plan for racial partnership in all its details vitally depends upon the co-operation of educated people of all kinds. The significance of education, and particularly the higher education of Africans, in the working out of such a plan must be clear to all.

F. G. YOUNG

ADMINISTRATION OF UNIVERSITIES

THE chuckles in academic circles when Prof. C. Northcote Parkinson presented his famous law to explain certain empire-building propensities of industrial leaders may now be echoed by chortles from industrial leaders who read an article by Parkinson which deals with related matters in universities (*The Universities Review*, 32, No. 1; October 1959).

Although British universities do not yet boast the organizational luxuriance of American universities—the University of Pittsburg has a chancellor, three vice-chancellors, three assistant chancellors and sixteen directors, eighteen deans and a host of other executives—Parkinson asks whether British universities are not proceeding in the same direction.

A recent study of the expenditure of some fifteen British universities shows that the percentage of their allocation to 'administration' varies between 5 and 10·6 per cent of their annual budget, Bristol spending the least and Exeter bracketed with

Leicester in spending the most. The average is about 7·2 per cent—a not manifestly exorbitant proportion. Expenditure between universities is not an easy comparison to make, for much depends upon the detailed layout of the estimates. Nevertheless, the universities which spend more than 10 per cent might learn something from those which spend only 5 per cent. What these figures do not reveal is the extent to which academic time and effort are diverted to administration on a part-time basis. This is a field in which statistical comparison is more difficult. All that is gathered is a vague impression of middle-aged scholars bewailing the administrative work thrust upon them; a complaint sometimes intended to excuse their lack of literary or scientific output. Eagerness for office is due to several causes, among which the most important is the least noticed. Presiding over a committee or dictating a routine correspondence is far easier than doing a good piece of research. It may often be better paid and frequently leads to the higher offices and honours, so that the scholar is offered every inducement to desert the library and take to the filing cabinets. Part of the trouble with British university administration centres on this inducement. For those most easily tempted along the easier path are not necessarily the most competent, though many of them prove to be so. In some instances, the burdens of which they complain have become burdensome as a direct result of their own inefficiency.

Those who admire the system of some larger American universities would wish to give more extensive powers to the executive head. There is no assurance, however, that this would be either an improvement or an economy. What is needed is an equivalent of the army 'p.s.c.', a higher qualification to be obtained by a short course and accompanied by an impartial and secret assessment. "Could not some university organize a staff course in higher administration, to be supervised by retired vice-chancellors and held at some centre for research which could be furnished with all the information available about all the universities of the world?" The course would be held in the vacation and designed for those who had been in charge of departments and for whom higher responsibilities might be predicted. After some experience of the course, universities might limit their higher offices to those who had gained this qualification.

For a scheme of this sort one strong argument from army experience is worth noting. Such faults as were apparent in the British generals of the Second World War were seldom or never attributed to lack of brains. There were failures in leadership; but the staff college system prevented any repetition of Balaclava. The same cannot be said of the university world. Such a staff college would provide a ready-made group of advisers to which a university might turn for guidance.

There are some arguments on the other side. Some of these will derive from that strange reluctance which academic persons reveal when invited to apply scientific method to university affairs. There is no insoluble problem in academic administration so long as the university is kept down to a reasonable size. But there are problems which remain unsolved merely because those immediately concerned lack the ability to find a solution and because they fail to apply the sort of method which they would apply without hesitation to matters more obviously scientific. In this matter of using a methodical approach to a political problem, the universities have not only a duty to themselves but also a further duty, by way of example, to the world.

THE NUFFIELD FOUNDATION

THE fourteenth annual report of the Nuffield Foundation (pp. xii+201. London: The Nuffield Foundation, 1959), covering the year ended March 31, 1959, records for the first time expenditure on grants exceeding £1 million, and of this £1,137,900, roughly a quarter was spent in the Commonwealth, mostly in Africa, and the rest at home on scientific, medical and social projects and on fellowships With regard to the last it may be noted that since 1946, 357 travelling fellowships have been awarded, and from the beginning the Foundation has made a special point of enabling the married overseas fellow to be accompanied by his wife and for its married Fellows has provided a small colony of furnished flats in Prince Albert Road, London. Travel grants during the year have covered the visit to England of Mr. R. C. Robinson, a neurosurgeon from the University of Otago Medical School; a visit to Dr. D. G. Abraham, Ibadan, by Dr. Wallace Brigden to study a puzzling heart condition; a lecture tour in Canada by Sir Christopher Ingold, arranged by the Chemical Institute of Canada; and a study visit on Commonwealth constitutional law by Prof. C. H. Alexandrowicz, of the University of Madras. Three medical fellowships, three fellowships and four scholarships in dentistry were awarded during the year, but the scheme for sociological scholarships and bursaries was discontinued and a new scheme of senior sociological scholarships initiated experimentally for three years. Only one travelling fellowship for Home Civil servants was awarded, but eight travelling fellowships were awarded in medicine, twelve in natural science, and nine in humanities and social science, while twenty-three awards were made under the Royal Society–Nuffield Foundation Commonwealth bursaries scheme.

In this report the Foundation, welcoming the new emphasis placed by the Government on the teaching of technology, records that, after considering how best it could help without trespassing on public responsibilities, the Foundation has decided that its chief interest should be in the technological colleges providing advanced courses. These colleges have a different purpose from the technological departments of universities, deriving from the belief that some students mature better if their theoretical training is more closely interwoven with industrial practice than is usual or desirable for university students. It considers that all such colleges need to establish a fruitful partnership with the industries with which they are most intimately associated, and while refraining from adopting a detailed programme of schemes for sandwich courses, for example, for which it would be willing to provide grant-aid, has indicated its interest in experimental schemes designed to achieve closer unity between the colleges and industry and in schemes which would benefit technological industry generally. As regards support for research at Commonwealth universities and institutions of like standing, the emphasis during the year has been on Africa.

In science, three grants of £50,000 were made during the year: one to the University of Edinburgh for fundamental research under Profs. M. Swann and C. H. Waddington on fibroblasts which may throw some light on the causes of rheumatism; another to the British Museum (Natural History) for the purchase of about half the collection of meteorites formed by Mr. H. H. Niniger, of Arizona; and the third, over five years, to the University of Cambridge for the development by Dr. M. V. Wilkes, of the Mathematical Laboratory, of a much faster digital computer, *Edsac* 3, capable of solving partial differential equations. Among other grants may be noted a further £9,000 over three years in support of the study of protein metabolism and particularly connective tissue under Prof. A. Neuberger at St. Mary's Hospital Medical School; £4,350 over three years for an investigation by Dr. I. Rorison, of the University of Nottingham School of Agriculture, of the effects of aluminium on plant growth; £11,000 over three years for the biochemical and physiological investigation of cell division and expansion in the meristems of the shoot and root at the University of Edinburgh; and a further £5,250 over three years to enable Dr. A. G. M. Weddell to continue his studies at the University of Oxford on the mechanisms of common sensation; and £1,690 a year for three years to King's College, London, for an investigation of the cell cortex and cell movement under Prof. J. F. Danielli.

In technology a research unit for psychological and social research on problems of technical education and human factors in industry has been established at Brunel College of Technology, Acton, with the help of a Foundation grant of £15,000, and £12,000 over five years was also offered to the University of Birmingham to establish a research unit in the Institute of Education under Prof. E. A. Peel to study and advise on the placing, counselling and teaching of part-time technical college students.

Apart from the grants to the University of Edinburgh for work on fibroblasts and the nature of the enzyme systems involved, all other new grants for research in rheumatism have been made to departments or projects already supported by the Foundation. These included £3,000 over three years for an electron microscope study at the Rheumatism Research Centre, University of Manchester, of the fine structure of the fibroblast in different phases of its metabolic activity, and £4,100 to the University of Sheffield for work under Dr. H. F. West in the Sheffield Centre for the Investigation and Treatment of Rheumatic Diseases. Other grants for medical research have included £25,000 over five years for a metabolic research unit at the Institute of Urology, University of London, and £20,000 towards the capital expenditure at Banstead Place and Dorincourt of two units for the rehabilitation of young chronic sick, chiefly for building and equipping a physiotherapy unit at Banstead Place.

Grants in the United Kingdom for social research and experiment in education include one of £7,500 over three years to the School of Architecture, University of Cambridge, for studies of urban reconstruction, particularly of urban housing in relation to daylight, sunlight and open space, including the provision of special apparatus, such as an artificial sky and the necessary measuring instruments for the study of day-lighting. A grant of £4,000 for two years was made to the Christian Frontier Council in aid of a study of the notion of equality in modern society with special reference to its implications for politics, industry and education, and for the pursuit of the ideals of excellence; one of £2,200 to the University College of North Staffordshire to enable a study of Parliament on the lines of a pilot study completed for 1955–58 to be made for the period 1945–55; a grant of £1,250 to the Institute of Race Relations, London, for a study of the integration of alien or minority groups of workers (chiefly West Indians and Poles) in an industrial situation in the London area; one of £3,700 to the School of Social Studies, University of Sheffield, for an inquiry by Mr. R. R. Kelsall into wastage, after training, among women teachers in England and Wales. A grant of £2,000 was made to the University of Manchester to enable Prof. S. Devons to try out his scheme of using honours graduates in physics part-time on research at a university and part-time on school teaching to relieve the shortage of first-class teachers of science in grammar schools, and £8,500 was offered to the Scottish Council for Research in Education to permit a follow-up survey between eleven and eighteen of the mental survey of the whole eleven-year age-group of more than 70,000 children in Scotland made in 1947, while up to £3,500 was offered to initiate "Youth Ventures", a non-profit making company intended to provide an unconventional youth service to fill in the gaps until the 'new towns' are more fully developed commercially.

In the field of research in ageing and care of old people, £6,000 was provided to the University of Cambridge for further studies of the economic position, and a grant of £3,700 to the University of Bristol for studies of the employment of older workers was renewed for a sixth and final year. The University of Birmingham received another £6,500 for Prof. P. L. Krohn's unit studying the ageing ovary, and the University of Leeds £6,000 for a further year's study by Dr. D. A. Hall's team of the changes taking place in connective tissue and the processes concerned in the degradation of such tissue during ageing, while £4,500 over the next three years was provided for work on physiological and pathological changes in the aged under Dr. E. Woodford-Williams at the Sunderland General Hospital.

Among grants for the Commonwealth Overseas during the year are £3,000 over two years for a study of the normal lymphatic system in typical primates, and £3,320 over two years for studies on the helminthic fauna of freshwater fish in Central Africa at the University College of Rhodesia and Nyasaland; up to £50,000 including a capital grant up to £30,000 over five years for an experiment in rural education by the Ministry of Health, Kenya; £5,000 to University College, Ibadan, in support of epidemiological field work in Nigeria; £3,000 as an emergency measure to extend the life of the Medical Research Unit, University of Natal; £20,000 over two years to the East African Agriculture and Forestry Research Organization for grassland reclamation in the Ngorongoro Crater highlands; a further £5,000 to the Gayaza Girls' High School for its study of farming and a balanced diet; a further £5,750 to the University College of East Africa for its swamp research; £13,850 for the study of urbanization in the Sierra Leone Protectorate being undertaken by the University of Edinburgh under Dr. K. L. Little; £12,000 for five years to the radio education unit of the University College of the West Indies; £20,000 over three years on a tapering basis to the Government Central Pedalogical Institute, Allahabad, for teaching English; up to £4,000 to the University of Hong Kong to enable Prof. J. E. Driver to purchase equipment for his Department of Chemistry, and £3,600 over three years to the Institute of Child Health, Calcutta, for studies of the pattern of disease in a sample group of children.

PROGRESS IN FOOD INVESTIGATION

FOR many years the annual report published by the Department of Scientific and Industrial Research under the title "Food Investigation" has reviewed the work of the Food Investigation Board and of three research institutes, namely, the Low Temperature Research Station at Cambridge, the Ditton Laboratory, Maidstone (with its associated unit at Covent Garden), and the Torry Research Station, Aberdeen.

In 1958 the Council of the Department of Scientific and Industrial Research decided on a series of administrative changes: (1) to disband the Food Investigation Board; (2) to retain the Torry Station as an independent establishment of the Department; (3) to transfer the other two institutes to the Agricultural Research Council. This latter change was brought about "because the two Councils believe that it is in the national interest to bring all Government research on food, other than fish, under one organization, the aim being to make the research more effective". From now onwards a report on the Torry Research Station will be published annually by the Department of Scientific and Industrial Research; the report, "Food Investigation 1958"*, is limited to the Cambridge and Ditton Laboratories and will be the last to appear under the auspices of the Department.

The Cambridge and Ditton Laboratories are complementary to one another: at Ditton the emphasis is on problems associated with fruits and vegetables; at Cambridge, although important work is being carried out on plant tissues and products, the major stress has been on animal products—meat (including bacon and poultry), eggs and animal fats. At both centres there have been fundamental investigations in biochemistry, biophysics and microbiology, and the report illustrates how difficult it is (in any event in the field of food technology) to draw a distinction between pure and applied research; the examination of the factors governing the quality of meat, eggs or fruit has stimulated basic work on the chemical composition of these materials and on the nature of the enzymes present; in turn, the newer information obtained has helped in the solution of practical problems of food storage and preservation.

The Cambridge section of the report includes summaries of investigations on three recent developments in food technology, namely, the administration of œstrogenic hormones to young animals (to increase the rate of gain of weight and the efficiency of food conversion), the use of antibiotics in poultry storage and of radiations in meat preservation. Details are given also of recent research on the phenolic constituents of plants; these compounds—ranging from relatively simple molecules to complex polymers—are now known to play a significant part in the control of colour, flavour and texture in a wide variety of foods.

The Ditton Laboratory records the increased commercial use of the hydrocooling process for vegetables such as watercress and lettuce; controlled experiments have shown that vegetables subjected to this process should arrive fresher on the market and retain their quality for a longer period. Another important line of research is concerned with the storage of potatoes in bulk using a new method of suppressing sprout growth—the introduction of nonyl alcohol vapour into the ventilation system of the store. An account is given also of various lines of research in plant physiology and biochemistry, and in refrigeration and other aspects of 'biological engineering'.

The food industries in the United Kingdom owe a debt of gratitude to the Department of Scientific and Industrial Research for sponsorship of research over some decades; the annual reports of the Food Investigation Board and other publications from the individual laboratories have played an indispensable part both in stimulating industrial research and in placing food storage and processing in the United Kingdom on sound scientific foundations.

FRANCIS AYLWARD

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[*] "Food Investigation 1958." Pp. vi+32. (London: H.M. Stationery Office, 1959.) 3*s*.

THE HOVERCRAFT

A DEVICE for forcing out compressed air between a ship's surface and the water was patented in 1883. It did not work. Many similar fruitless attempts were made in the next sixty years until, in 1953, Mr. C. S. Cockerell began experimenting with air lubrication. His success is now universally recognized and the first hovercraft has begun the hard task of practical development, foreshadowing a new contender in the field of maritime transport.

Cockerell began as had De Laval, by attempting to contain a sheet of air between the surface of a vessel, in this case a planing craft, and the sea. He found that the reduction in drag was offset by the hull modifications needed to supply the air. He then attempted to contain the air lubricating sheet between vertical plates but these plates increased the skin friction drag. The answer was to replace the solid containing plates by curtains of air and so the hovercraft was evolved.

The experimental version now flying, the 'SR-NI', is an oval dish on top of which are mounted the propulsion and control systems, and the air compressor. The compressor is a 7-ft. diameter ducted fan powered by a 435-h.p. internal combustion aero engine. Two-thirds of the compressed air is fed to two concentric annular slots near the periphery of the under surface of the oval disk. The slots are shaped so that two continuous jet sheets are directed downwards and slightly inwards. When the engine is started the jets first build up the air cushion under the disk and then maintain the pressure difference between the cushion and the atmosphere. The pressure difference deflects the air curtains and they spill outwards at the ground or water surface (the pressure difference times the height above the ground must equal the radial rate of change of momentum). Two curtains are used for stability reasons. A third of the compressed air is bled off into two parallel propulsion ducts of rectangular cross-section which lie along and above the flat sides of the oval disk. Vanes at each end of each duct provide the necessary longitudinal and lateral control.

Because the weight that can be supported is dependent on the disk area while the power required is proportional to the peripheral length, the economy of a hovercraft should improve with size. A reasonably flat operational surface is required and early applications may well be over surfaces which existing vehicles find difficult, for example, marsh, snow, sand, ice and shallow rivers. Serious consideration has been given to a 400-ton car ferry capable of carrying eight hundred passengers and eighty cars across the channel at a speed of 90 knots. A careful detailed study of hovercraft economics suggests a cost of between 8 and 18 pence per (payload) ton (nautical) mile in the speed-range 70–170 knots. Both cost and speed lie somewhere between the current values for ships and commercial aircraft. If the hovercraft can prove its worth with modest payloads over short distances, then serious thought could be given to developing a trans-ocean hovercraft capable of crossing the Atlantic in 24 hr. This new competitor could not appear within the next ten years, however, and one wonders what the level of ship and particularly aircraft fares will be by then. Nevertheless, the hovercraft has an extremely interesting future and one can only congratulate the inventor and the development engineers on their achievement and remark that this is one enterprise where the Old World leads the New.

These remarks are based on a paper "The Hovercraft—a New Concept in Maritime Transport", by P. R. Crewe and W. J. Eggington, read before the Royal Institution of Naval Architects on November 19, 1959. J. L. STOLLERY

NEW DELHI SYMPOSIUM ON ALGAE

POSSIBLY because algae are usually inconspicuous in the temperate regions in which most microbiologists receive their training, the dominant role which these organisms may have in soil and water in other climates is apt to be overlooked. In India, although much excellent work has been done on algae, this has largely followed morphological and systematic lines, and relatively little attention has been paid to their activities and possible economic importance. The symposium on "Algology", held in the Indian Agricultural Research Institute, New Delhi, during December 7–11, under the auspices of Unesco and the Indian Council of Agricultural Research, had the object of stimulating investigations on the more practical aspects of the subject. It was opened by Mr. S. K. Patil, Minister for Food and Agriculture, and was attended by experts from Japan, England, the Federal Republic of Germany, and Israel, as well as by delegates from all parts of India.

An introductory address by Dr. M. S. Randhawa, chairman of the organizing committee, outlined the development of algal studies and lines of research which may be of practical importance. The principal uses to which these organisms may be put are mentioned below, but it may be noted that in the tropics algal growth often needs controlling rather than encouraging—a matter which was not considered in any of the papers which followed. The growth of blue-green algae, which generally occurs on buildings during wet weather, not only renders them unsightly but also corrodes the stone-work and presents a problem of major economic importance. Algal growth may also interfere in the manufacture of salt, it may block canals and irrigation channels, and pollute fresh water.

The papers presented covered nearly every aspect of modern knowledge of the algae. Classical morphology was represented by papers on the comparative morphology of the Charophytes, by V. S. Sundaralingam, and on reproduction in red algae, by M. S. Balakrishnan, and by a lecture by M. O. P. Iyengar, describing some of the many interesting species which have been discovered in India. Recent advances in the study of the submicroscopic morphology of algae were reviewed by T. V. Desikachary, who also discussed the structure of diatom frustules in the light of his own electron microscope investigations. Papers on algal cytology, which is especially important for the understanding of life-cycles and for taxonomy, were given by C. S. P. Rao, Y. S. R. K. Sarma and Y. B. K. Chowdary.

One of the most important of recent advances in algal physiology has been the introduction by H. Tamiya and his associates of the technique of synchronous culture as a means of studying the metabolic changes occurring during the life-cycle. A paper by H. Tamiya summarized the main results obtained and dealt particularly with the role of sulphur in the processes of cell maturation and division. Studies on the mineral nutrition of a *Selenastrum* were described by K. V. Natarajan. In a lecture on the extracellular products of algae, G. E. Fogg presented evidence showing that the liberation of these is widespread and that enzymes are included among the very varied kinds of substances involved. These extracellular substances may be of ecological importance in various ways, as, for example, by forming chemical complexes with inorganic ions. The kinetics of the production of extracellular nitrogenous substances by a nitrogen-fixing *Nostoc* isolated from the root nodules of *Trifolium alexandrinum* was discussed by G. S. Venkataraman. In another paper on nitrogen fixation, G. E. Fogg emphasized the close intermeshing of this process with photosynthesis which occurs in blue-green algae. He also reported nitrogen fixation by *Chlorogloea fritschii*, evidently the first unicellular member of the group in which this property has been demonstrated unequivocally. A paper by A. Watanabe dealt with the importance of blue-green algae in rice-fields. Nitrogen-fixing blue-green algae are found almost ubiquitously throughout tropical, sub-tropical and temperate regions and evidently contribute materially to the fertility of many soils. It has been found possible to grow *Tolypothrix tenuis*, a particularly vigorous nitrogen-fixing species, on the large scale and to preserve it as a culture on porous gravel. Inoculation of paddy fields with this has resulted in appreciable and long-continued improvements in yields of rice.

H. Tamiya, H. von Witsch and A. M. Mayer each considered various aspects of the mass culture of microscopic algae such as *Chlorella* for use as animal or human food. The average yields of *Chlorella* grown on the large scale correspond to the production of 14,000 lb. of protein per acre per year, whereas the corresponding figures are 600 lb. for grass, 420 lb. for peanuts, 90 lb. for milk and 54 lb. for meat. The results of feeding tests demonstrate that the alga is of high nutritive value. Convincing evidence of the feasibility of large-scale culture was given in a film of the work of the Japan Micro-algae Research Institute. This has culture ponds of about an acre in total area producing 12 tons dry weight of *Chlorella* a year, which is largely used as a source of growth factors for the commercial culture of *Lactobacillus*. As a means of producing material which can be used directly in human or animal nutrition the process is not yet an economic one; the estimated cost of decolorized algal powder is 1·12 dollars per lb.—considerably more than that of cheap protein sources such as soy beans (0·06 dollar per lb.) or fish meal (0·13 dollar per lb.) but a little less than that of skim milk powder (1·20 dollars per lb.). In arid regions this high cost may be offset by the high degree of water conservation which is possible with algal cultures as compared with conventional crops, but for more general use cheaper methods are essential. To suit Indian conditions, it would be desirable to simplify the process so that it might be operated by peasants, and to this end M. O. P. Iyengar suggested that an examination should be made of algae known to grow prolifically in Indian fresh waters and which might be harvested easily.

The cost of algal culture can be minimized by combining it with the photosynthetic treatment of sewage and organic wastes by the oxidation pond method. A paper submitted from the Sanitary Engineering Laboratory of the University of California gave a résumé of this process and reported that algae harvested from oxidation ponds by alum flocculation proved satisfactory as a protein source in the diet of lambs. Field studies carried out at the Engineering College, Guindy (Madras), on the use of algae in sewage disposal, described by A. Raman, indicate that the method can be used under Indian conditions.

In a general lecture on the use of algae as food, H. Tamiya, besides describing the possibilities of *Chlorella*, gave an account of the use of seaweeds for this purpose in Japan. Among these, the most popular is *Porphyra tenera*, which is cultivated in shallow seas to the extent of 4,000–5,000 tons dry weight per annum and which can command prices of up to 8.00 dollars per lb. The amounts of marine algae available around India are relatively small, but a paper by F. Thivy showed that there are, nevertheless, possibilities for their use as food and described cottage industry methods for the preparation of seaweed meal and agar. The ecology and distribution of Indian marine algae were considered in papers by J. N. Misra and K. S. Srinivasan.

Phytoplankton ecology was discussed in four papers. V. P. Singh described detailed observations made over a year on two lakes in Uttar Pradesh, the plankton of which is dominated by *Microcystis*. M. T. Philipose and S. V. Ganapathi gave more general descriptions of Indian freshwater phytoplankton in relation to fisheries and water supply. The marine phytoplankton of the west coast of India was considered in special relation to fisheries by R. Subrahmanyan. The indications are that the phytoplankton production could support a considerably higher rate of fishing than it does at present.

At the final session a number of recommendations were approved for the extension of algal research in India on a broad front with the ultimate object of making more practical use of these organisms.

G. E. Fogg

EROSION OF METEORITES IN SPACE AND THE DENSITY OF INTERPLANETARY GAS

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THE etching rate, due to interaction with interplanetary gas, for the Sikhote-Alin meteorite fall has been estimated[1] as $\lesssim 2 \times 10^{-7}$ cm./yr. from an exposure life-time of 5×10^{8} years, as determined by argon-38/argon-39 age measurements. From this result, Whipple[2] has suggested that the maximum possible proton density of interplanetary space in the vicinity of the Earth is as shown in column 3 of Table 1. Whipple assumes that meteoritic etching is due solely to sputtering from solar protons and uses normal incidence sputtering yields for protons on iron as follows: 0·5 atom/proton for 10,000 eV. or more, 0·2 at 500 eV. and $\sim$0·02 at 100 eV. In the actual calculation, these yields are doubled to account, approximately, for the greater yields which are manifest at smaller angles of incidence.

The purpose of this article is to point out that solar protons are probably not the major factor in determining etching rates from sputtering by gas clouds ejected from the Sun. Calculated upper limits on interplanetary gas densities and erosion rates near the Earth may be changed appreciably as a consequence of including sputtering by elements heavier than hydrogen, and are uncertain for other reasons as discussed below.

In spite of the lower abundance of helium relative to hydrogen, the greater momentum transfer and sputtering efficiency for this ion dominate the meteoritic sputtering process under the assumptions: (1) the ejected solar gas has a chemical composition approximately duplicating that of the solar corona; (2) significant fractionation of helium does not occur in the solar acceleration and escape processes. Since solar streams appear to be "segments of the corona, blown out bodily"[3], these assumptions do not appear unreasonable. In this connexion, it should be noted that Shklovskii's discussion[4] of sources for helium in the terrestrial atmosphere gives strong support to the view that the stream composition is similar to that of the Sun itself. If helium ions travel along in the ejected clouds with the same velocities (column 2 of Table 1) as the stream protons, they will, of course, have four times as much energy, and their relative flux will be the same as their relative abundance. The atomic abundances of hydrogen and helium in the solar atmosphere are difficult to determine. Neven and DeJager[5] suggest an hydrogen/helium ratio (δ) of 20/1 while Unsold[5] finds 5/1. Recently, Aller[6] has given 7/1 as the best value currently available.

Sputtering yields do not appear sensitive to energy for either hydrogen or helium ions in the range 1–10 keV. according to recent measurements by Grønlund and Moore[7]. These data, obtained with analysed beams known to contain only ions, refer to silver targets, but iron is expected to behave in a very similar manner. The ratio of helium ion sputtering efficiency to hydrogen sputtering efficiency (β) is roughly fourteen over the entire range studied. We shall compare hydrogen and helium yields at the same energy, thereby implicitly allowing for somewhat reduced acceleration efficiency for the heavier ions, and shall assume that the relative sputtering efficiency will not change radically when yields are integrated over all angles of incidence. Using the appropriate abundance and allowing for differences in sputtering efficiency, it is seen that the helium erosion is comparable to, and probably exceeds, that due to hydrogen, implying that Whipple's upper limit to the solar proton density can be markedly reduced if the effect of helium is included. Specifically, if N'_p is the proton density calculated by Whipple, one may write for the corrected proton density N_p:

$$N_p = N'_p\left(1 + \frac{\beta}{\delta}\right)^{-1}$$

Here I have retained Whipple's assumptions in allowing for angular dependence of yield (a factor of 2), absolute sputtering yield for protons (as listed earlier) and scaling from fluxes at the meteorite orbit to those near the Earth (a factor of 3/2). Results calculated from this expression, using $\delta = 7$ and $\beta = 14$, are shown in column 4 of Table 1. Including the high Z elements other than helium would reduce N_p still further, but unless their sputtering efficiency is surprisingly high, the change would not be large.

The figures of column 4 are low enough (especially at the higher velocities which, on the basis of other evidence, seem to be most probable) that one becomes concerned about maintaining explanations of a variety of other astrophysical and geophysical phenomena, which, in the past, have led to 'conventional' solar gas densities (at 1 A.U.) between 10^2 and 10^3 protons/cm.^{3} under quiet conditions with estimated velocities varying upward from 500 km./sec. Furthermore, Russian observations[8], while unfortunately not providing velocity information, have recently yielded measured densities which also lie between 10^2 and 10^3 protons/cm.^{3}. Recent laboratory sputtering results using analysed beams are of especial interest in this situation since they are markedly lower than the figures employed by Whipple which were obtained by Wehner using gas-discharge techniques. If one assumes that absolute yields of the order of those observed by Grønlund and Moore for protons on silver (0·035 at 5,000 eV.) are also appropriate for iron, the allowable proton densities rise markedly as shown in column 5 and come into conformity with estimates from phenomena other than meteoritic erosion. It may be that resolution of the somewhat conflicting requirements on fast (for example, >500 km./sec.) solar stream densities implied by meteoritic life-times and other types of observations lies, at least in part, in the reduced sputtering by beams containing only atomic ions. Completely ionized beams would be the rule in space, but perhaps have not been attained in most laboratory sputtering experiments. On the other hand, if the average solar proton velocities prove to be 300 km./sec. or less, one need not appeal to explanations of the sort suggested here.

Table 1. PROTON DENSITIES NEAR EARTH AS INFERRED FROM METEORITIC EROSION RATES

(1) Proton energy eV	(2) Proton velocity km./sec.	(3) Number* per cm.^{3}	(4) Number† per cm.^{3}	(5) Number‡ per cm.^{3}
500	310	290	95	1,000–1,500§
1,000	440	140	45	650
5,000	980	40	13	186

* After Whipple.
† Corrected for sputtering by helium.
‡ Corrected for sputtering by helium and using analysed beam sputtering yields.
§ Rough estimate using $\beta = 14$, which may not hold for this energy.

Finally, it should be noted that any of the results given in Table 1 must be viewed with caution for a number of reasons beyond those already discussed:

(1) The meteoritic etching rate employed at the outset may refer to heliocentric orbits at relatively high angles to the ecliptic and therefore may be anomalously low. Many meteors have been observed in 60 degree orbits. It appears that the solar corona, and probably solar streams, are markedly more dense in the ecliptic[9]. Determination, from fall data, of the orbits of meteorites is difficult and generally unreliable[10], hence study of a statistically significant number of meteorites would be highly desirable before reaching final conclusions.

(2) Energy transfer from the ions to the electrons in the region of solar stream instability may occur[11] and might be accompanied by a spreading of the solar streams away from the ecliptic, resulting in reduced particle densities. If the admittedly speculative transfer process is effective, some shift in the particle energy spectrum would be produced in the region beyond 1–2 A.U. Thus, in the asteroid belt, where the meteorite may have spent the greater part of its exposure life-time, it would be bombarded by an electron spectrum of higher energy and a proton spectrum of lower energy than that characteristic of the inner solar system. Decreased sputtering would result, so the relative gas densities and erosion-rates near the Earth would increase.

(3) If the meteorite originated in the asteroid belt and was deflected into an Earth-capture orbit in a

collision, it may have spent some of its life well beyond 2 A.U., since most asteroids have mean distances from the Sun lying between 2·0 and 3·5 A.U., but are found over a much wider range. Other than by statistical arguments, it does not seem possible to state definitely that the meteorite spent most of its life in an unperturbed Earth-capture orbit with orbital elements near those of the Earth itself. Thus, a square-law dependence of solar stream densities could introduce appreciable uncertainty in scaling from the meteorite orbit to a circular orbit of 1 A.U. radius.

Direct measurement of both erosion rates on known surfaces, as well as particle densities and velocities in space near the Earth, would obviously be of great value, as would collection and analysis of additional data on meteoritic etching rates. Experiments are planned in various laboratories to make direct observations on interplanetary proton densities. In the present context, it would seem desirable to attempt simultaneous measurements on helium.

[1] Whipple, F. L., and Fireman, E. L., *Nature*, **183**, 1315 (1959).

[2] Whipple, F. L., *J. Geophys. Res.*, **64**, 1655 (1959).

[3] Keipenheuer, K. O., "The Sun", ed. G. P. Kuiper, **1**, 449 (University of Chicago Press, 1953).

[4] Shklovskii, I. S., *Soviet Astronomy*, **2**, 516 (1958).

[5] Quoted by Aller, L. H., in "Handbuch der Physik, Astrophysics II Stellar Structure", **52**, 344 (Springer Verlag, Berlin, 1958).

[6] Aller, L. H. (private communication, January 5, 1960).

[7] Grønlund, F., and Moore, W. J., *Science*, **30**, 1417 (1959); *J. Chem. Phys.* (in the press). I am indebted to Dr. Moore for communication of his results prior to publication, as well as helpful comments on the relative behaviour of iron and silver.

[8] Sedov, L. I., and Krassovsky, V. I. (private communication, November 20, 1959).

[9] See discussion, Symposium on Exploration of Space, *J. Geophys. Res.*, **64**, 1698 (1959).

[10] Urey, H. C., see discussion, Symposium on Exploration of Space, *J. Geophys. Res.*, **64**, 1733 (1959).

[11] Reiffel, L., *Nature*, [**185**, 229 (1960)].

DIETARY PHYTATE AS A POSSIBLE CAUSE OF MAGNESIUM DEFICIENCY

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WE have recently made a study in the rat of the effects of dietary phytate upon intestinal phytase and bone calcification. A short account of some of the results has been published[1].

During the course of these observations, we found that rats given sodium phytate in purified diets became severely ill. We believe that this was due to a conditioned deficiency of magnesium, brought about by the soluble phytate combining with the dietary magnesium, which consequently becomes insoluble and unavailable.

The basal purified diet was made up of sucrose 60, low-vitamin casein 20, arachis oil 15, salt mixture 5. Additions of sodium phytate (Ciba) were made at the expense of sucrose. Vitamin supplements were given separately. Most of the experiments were carried out with a strain of albino rats; in a few experiments we used hooded rats of the Lister strain, and obtained similar results, though this strain appeared somewhat less sensitive to phytate. This recalls the observation on chronic magnesium deficiency in two similar strains, described by Watchorn and McCance[2].

We observed the signs of phytate intoxication in albino rats with levels ranging from 1 to 10 per cent sodium phytate in the diet. The animals were put on the experimental diet at 24 days, when they weighed about 45 gm. At a level of 10 per cent phytate, the rats developed severe diarrhœa within 24 hr. and rapidly lost weight. Within a week, seven out of the eight rats in this group had died, having lost about 10 gm. body-weight. The last rat survived for four weeks, but at the end of that time weighed 64 gm., about one half the weight of the control animals receiving no phytate.

With 7½ per cent sodium phytate, severe diarrhœa again developed in the first day, but subsided during the second week. The rats lost weight during the first week, then began to put it on again. They were nevertheless much thinner than normal, with a coarse staring coat and bald patches. The nose was swollen and covered with a dark red bloody exudate. The abdominal wall was hard, and there was often priapism. The animals were very irritable and difficult to handle. They died after about 18 days on the experiment, weighing about 80 gm., with evidence of having passed blood rectally.

With 5 per cent sodium phytate, diarrhœa also occurred, but less severely and rather later. The rats gained weight slowly after the first week. They also developed swollen noses with exudate. One out of the four animals had a convulsive fit in the third week, and showed paralysis from the waist downwards. It lay on its side, breathed very rapidly and died within 24 hr. The others died after about 25 days, weighing about 105 gm. Hooded rats showed an exactly similar response to 5 per cent phytate.

With 2½ per cent sodium phytate, there was only slight diarrhœa in the first week. Bloody exudates again appeared on the noses of the rats. Two out of four rats had convulsive fits and developed paralysis a few days before death. All of them died after about 50 days, weighing about 150 gm.

With 1 per cent sodium phytate, there was no diarrhœa. The bloody nasal exudate appeared in two out of four rats. Growth was better, though still less than that of the control animals receiving no phytate. One rat was found dead after five weeks, and one after eight weeks; there was no sign of convulsions or of paralysis. The other two rats survived at least nine weeks. On this level of 1 per cent sodium phytate, 16 hooded rats showed no signs other than diminished growth.

At first, we examined the possibility that the signs were due to deficiency either of calcium or of inositol. Calcium deficiency could arise by the phytate making the dietary calcium unavailable. We did not think that calcium deficiency was very likely; while it might perhaps account for the convulsions and paralysis, the other signs have not been described in calcium deficiency. Moreover, the toxicity produced by phytate did not occur with dietary citrate, which like phytate would have rendered calcium unavailable. Nevertheless, we tried to cure or to prevent phytate toxicity by the addition of calcium to diets containing phytate, or by the parenteral injection of calcium gluco-galacto-gluconate ('calcium Sandoz'). We found no difference in the signs produced by phytate when the rats were given calcium.

Inositol deficiency could arise by competitive inhibition, since phytate is inositol hexaphosphate. Moreover, the signs we observed were similar to those which had been reported for inositol deficiency in rats. We therefore gave 50 mgm. inositol daily, in addition to the 1·1 mgm. contained in the routine vitamin supplements, to half of a number of rats receiving varying levels of phytate from the age of 24 days. This made no difference in growth or in the development of signs of phytate intoxication.

We now examined the possibility that phytate intoxication was due to magnesium deficiency. We recalled that Kruse, Orent and McCollum[2] and Watchorn and McCance[3] had reported signs of magnesium deficiency in rats, which resembled those in our rats showing phytate intoxication. We therefore gave levels of phytate up to 7½ per cent to groups of eight animals; four on each level had in addition 10 gm. magnesium sulphate to 100 gm. diet. The additions were made after the animals had been on the control or phytate diets for three weeks, or (with 7½ per cent phytate) two weeks. The general effect of the magnesium supplements was to improve the appearance of the animals; for example, the nasal exudates were much reduced, and there were no bald patches. There was no effect on the growth of the animals receiving 1 per cent phytate, but there was improved growth and longer survival in those receiving 2½, 5 or 7½ per cent phytate. In the 5 per cent group, for example, the rats receiving magnesium gained on average 20 gm. in weight in the week following the addition, whereas those not receiving the magnesium gained 8 gm.; the rats receiving magnesium survived at least 56 days, whereas those not receiving it died after an average of 41 days.

In this experiment, there were two animals which showed exceptional responses. One was receiving a diet with 1 per cent sodium phytate; it was also given magnesium supplements, but in spite of this it died in convulsions. The second animal was receiving a diet with 7½ per cent phytate; it was not given magnesium supplements, yet nevertheless it showed no signs of phytate intoxication throughout the experiments. These great variations between individuals in the development of signs of hypomagnesæmia are well known in cattle (Allcroft, R., personal communication). Apart from these individuals, however, all the rats receiving 2½ per cent or more of phytate without magnesium were either found dead, or developed fits and died within 24 hr.; all the rats receiving the same diets with magnesium gained weight and survived for the duration of the experiment and showed little if any signs of intoxication.

A second experiment was carried out with five groups, each of eight rats. One group received no phytate, two groups received 5 per cent phytate and two groups 7½ per cent phytate. One group on each level of phytate was given in addition magnesium sulphate at the rate of 10 gm. per 100 gm. diet. The rats on the phytate-free diet, or on the phytate diets with magnesium sulphate, all grew and survived until the experiment was terminated after nine weeks. Those receiving 5 or 7½ per cent phytate with no magnesium grew badly, and died on average at 42 days or 20 days (see also Fig. 1). Although the addition of magnesium greatly improved the condition of the animals receiving phytate, their rate of growth was lower than that of the controls receiving no phytate.

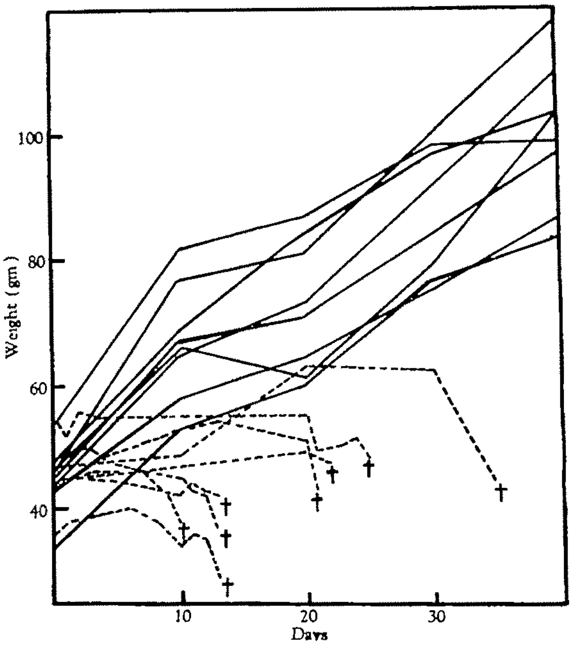

Fig. 1. Effect of addition of magnesium to a diet containing sodium phytate

All rats were given a purified diet with 7·5 per cent sodium phytate from age 24 days Curves represent weight changes in individual rats. ——, diet with 10 per cent magnesium sulphate; – – –, diet without magnesium sulphate

Since our work was primarily designed to study quite a different problem, we have not yet pursued the many questions which are raised by these results. In particular, we have not examined levels of magnesium in the blood. Nor have we at present facilities for studying the condition in ruminants. We believe, however, that we have reasonable circumstantial evidence that diets which include phytate may cause magnesium deficiency.

We may summarize this evidence as follows: (1) soluble phytate can form insoluble mixed salts of calcium and magnesium; (2) the signs of intoxication shown by rats given diets rich in phytate resemble those described in rats given diets deficient in magnesium; (3) rats show a varying individual susceptibility to phytate intoxication, recalling the known variability of signs of magnesium deficiency in cattle; (4) the signs of phytate intoxication are to a great extent prevented or cured by the administration of magnesium; (5) the doses of magnesium needed to prevent or cure the signs of phytate intoxication in rats are surprisingly large, again recalling the situation in hypomagnesæmia in ruminants.

We suggest, therefore, that a dietary constituent such as phytate, which can render magnesium unavailable, may be a cause of hypomagnesæmia in adult ruminants. It is likely, for example, that this is the explanation of the results of McAleese and Forbes[4], who showed that a diet high in roughage can cause hypomagnesæmia in lambs. The roughage included corn cobs, and corn cobs were also used for bedding. Although we do not know how much phytate was consumed, and how much of it was soluble phytate, it is likely that the small amount of dietary magnesium was still further reduced by combination with soluble phytate.

Our hypothesis could also explain why the level of serum calcium is often decreased in hypomagnesæmia in adult cattle[5]. Substances such as soluble phytate form insoluble salts with calcium as well as magnesium, so that excess of dietary phytate would lead to decreased absorption of both.

The confirmation of our hypothesis requires much further work. In particular, we need to know the effect of dietary phytate on the magnesium and calcium levels of blood, and whether the pastures which cause hypomagnesæmia in ruminants contain high amounts of phytate or some other constituent which binds the magnesium.

[1] Roberts, A. H., and Yudkin, J., *Proc. Nut. Soc.*, 18, 27 (1959).
[2] Watchorn, E., and McCance, R. A., *Biochem. J.*, 31, 1379 (1937).
[3] Kruse, H. D., Orent, E. R., and McCollum, E. V., *J. Biol. Chem.*, 961, 519 (1932).
[4] McAleese, D. M., and Forbes, R. M., *Nature*, 184, 2025 (1959).
[5] Allcroft, W. M., *Vet. J.*, 103, 2 (1947).

FLUORESCENCE STUDIES OF CHLOROPLAST DEVELOPMENT IN *EUGLENA*

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EUGLENA should be an excellent organism for the study of control and biosynthesis of a new organelle in response to a highly controllable and defined external stimulus because the chlorophyll-free and presumably chloroplast-less dark-grown organism is believed to develop both rapidly after being placed in the light. An early electron microscope study[1] of the transition seemed to show no detectable chloroplasts in dark-grown cells. As pointed out by Nishimura and Huzisige[2] the results of that work are questionable because Wolken and Palade reported the presence of a very considerable amount of chlorophyll in their dark-grown cells whereas in cells grown for many weeks in the dark there is no trace of chlorophyll[2,3]. Indeed, with so much chlorophyll present, it would be expected that some chloroplast remnants would have been found.

The fact that chlorophyll has a strong red fluorescence suggested to us that it might be possible to follow the developing plastids by direct observation with the light microscope under conditions of dark-field illumination. A similar technique has been used by Strugger[4] and by Heitz and Maly[5] for the chloroplasts of higher plants.

Light from a mercury arc was filtered to pass only 435 mμ; a yellow filter in the eyepiece eliminated interference from the light source. A preliminary look at light-grown cells revealed a spirally arranged set of red-fluorescing structures with the typical disk shape of chloroplasts. A similar look at dark-grown organisms revealed a spirally arranged set of light pink structures scarcely larger than points.

The following technique was developed to immobilize the cells for photography. A 5 per cent solution of polyvinyl alcohol in distilled water is prepared by boiling lightly and letting it stand overnight at room temperature to ensure that the alcohol is fully dissolved. A few drops are placed on a clean glass microscope slide and spread over the entire slide; these dry face up for at least 2 hr. at 37° C. A cover should be placed over the slides to prevent dust from falling on them. We find such slides still usable after as many as 3 days in the incubator. Gelatin and agar were tried and found to have too great fluorescences of their own.

The dried slide is placed in observation position in the fluorescence microscope. A small droplet of the solution to be studied is placed on a clean cover-slip which is inverted on to the slide. The droplet spreads and is rapidly absorbed by the alcohol layer, immobilizing the cells. Depending on the stage of chloroplast development, there are 2–10 min. for making pictures before observable deterioration is found. Both 'Ektachrome' and 'Superanschchrome' daylight films have been used, though the former has some advantages for this system because of its greater response in the red.

Euglena gracilis var. *bacillaris* was cultured in Hutner's *p*H 3·5 medium as previously described[6]. At zero time, several ml. of dark-grown logarithmic-phase cells were placed in a test-tube in the light of a daylight fluorescent lamp at room temperature. Samples were studied at various times after the cells were placed in the light. The zero-time samples were taken directly from the culture growing in the dark.

At zero time, the cells usually have one or two pinkish bodies in a line of smaller, paler dots of fluorescence usually running spirally along the organism. The organism itself has a bluish fluorescence; the flagellum is light green. By 15 min., the small dots are visibly enlarged and have become orangish. By 1 hr. the dots are orange-red objects about one-fifth the length of a mature chloroplast. There is some indication that, after this time, some of the increase in size of the plastid may be due to coalescence of several smaller bodies.

By 4 hr., some of the plastids are as much as one-third normal length, and are now quite red. There is an appreciable inhomogeneity of development not only from organism to organism, but also among plastids of the same organism. At about this time, the blue fluorescence has begun to disappear and is essentially gone within another hour or so. Thereafter the plastids develop regularly, on the average, with occasional normal-sized chloroplasts being detectable after about 20 hr. in the light.

There appear to be roughly two types of mature structure. In the majority type there are quite normal-looking plastids with occasional hints of a line of red fluorescence running between some of the plastids. In the minority type the connecting lines are broadened so much as to present a ribbon-like structure with the chloroplasts being detectable only by their being more deeply coloured.

From these observations it may be concluded that there are what must be termed proplastids of an appreciable size (perhaps 1μ) even in the dark-grown organism. It also seems clear that the development involves an actual increase in plastid size as well as in complexity. Thus, it would not be true that the chlorophyll simply vanishes when a green cell is placed in the dark, leaving a structure which can later be refilled whenever chlorophyll synthesis is re-initiated. Preliminary observations with the electron microscope support this conclusion.

The frequent links between chloroplasts remain to be interpreted. Two interpretations may be proposed here. First, the entire chloroplast structure in *Euglena* is to be thought of as a single chloroplast with the normally designated chloroplasts being, in reality, more analogous to the grana of higher plants. Secondly, one might consider that the links are the remnants of connexions between dividing plastids.

Preliminary observations of the reverse processes occurring when light-grown cells are placed in the dark have revealed that the steps are the reverse of the synthetic steps, with, however, a much different time-scale. For example, the blue fluorescence reappears only after more than a day in the dark. The plastids in these organisms remain for many days, being reduced to about one-fifth normal size only after about 2 weeks.

A preliminary survey of our collection of mutants has shown appreciable differences even among different albino mutants. For example, one albino has an overall greenish colour; a second has pale yellow-green plastid-like bodies; and a third has no appreciable fluorescence. A yellow-brown mutant looks normal when grown in the dark, but when grown in the light contains structures similar to those found in normal cells after 10–15 hr. of light.

We have been able to use fluorescence as a criterion of purity of plastids being extracted from organisms at various times after being placed in the light. Also, it now appears likely that the fluorescence may be used as a criterion of functional integrity in such extracts.

Finally, there is one observation that can be made only with the freely moving organisms. The entire chloroplast structure does not move with respect to the cell surface, being apparently fixed in some manner to the cell wall or membrane.

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[1] Wolken, J. J., and Palade, G. E., *Ann. N.Y. Acad. Sci.*, 56, 873 (1953).

[2] Nishimura, M., and Huzisige, H., *J. Biochem.*, 46, 225 (1959).

[3] Huzisige, H., Terada, T., Nishimura, M., and Uemura, T., *Biol. J. Okayama Univ.*, 3, 209 (1957).

[4] Strugger, S., *Naturwiss.*, 37, 166 (1950).

[5] Heitz, E., and Maly, R., *Z. Naturforschung*, 8, *b*, 243 (1953).

[6] Lyman, H., Epstein, H. T., and Schiff, Jerome A., *J. Protozool.*, 6, 264 (1959).

INCORPORATION OF ORTHOPHOSPHATE LABELLED WITH PHOSPHORUS-32 INTO CEREBRAL PHOSPHOLIPIDS *IN VIVO*

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ALTHOUGH it was originally considered that the rate of incorporation of radioactive phosphate into brain phospholipids *in vivo* was relatively slow, the work of Dawson and Richter[1] showed that the rate of incorporation of label from the trichloroacetic acid-extractable phosphorus compounds of cerebral tissues into phospholipid phosphorus was relatively high. Ansell and Dohmen[2] showed that the incorporation varied for different phospholipids over short time intervals (0–7 hr.), though the difficulties of interpretation are manifold[3]. Marinetti and co-workers have recently published time curves for the incorporation of phosphorus-32 into some of the phospholipids of tissues other than brain for periods of up to 100 hr. following the subcutaneous injection of labelled orthophosphate into adult rats[4]. We wish to record results showing the course of the incorporation of orthophosphate labelled with phosphorus-32 into brain phospholipids and two probable precursors, phosphorylcholine and phosphorylethanolamine, over a period of seven days; some information on the rate of incorporation of phosphorus-32 into the monoesters of brain *in vivo* has been recorded previously[5,6]. The present communication is prompted by a report by Miani and Bucciante who have carried out a comparable study[7].

Three litters of three-week-old rats were injected intraperitoneally with 4μc. labelled orthophosphate/gm. body-weight (supplied in sterile isotonic solution with carrier phosphate by the Radiochemical Centre, Amersham, Bucks) and killed at intervals. Phosphorylethanolamine and phosphorylcholine were separated from the trichloroacetic acid-soluble fraction as previously described[6]. Phospholipids were extracted from the washed tissue and the specific radioactivities of individual phospholipids determined by the method of Dawson[8] as described previously[9]. The results are recorded in Figs. 1 and 2 in which the specific radioactivities of the various fractions (all corrected to the same radioactive standard) are plotted against time.

The specific radioactivity of the acid-soluble fraction of brain tissue rapidly rose and then stayed relatively constant as has been noted by Dziewiatkowski and Bodian[10]; they found that its rate of decline was much slower than that of the blood. The present results (Fig. 1) indicate that phosphorylethanolamine and phosphorylcholine acquire the same radioactivity as the acid-soluble fraction as a whole within 24 hr. Previous observations[6] have shown that after an exchange period of 3 hr. the specific radioactivity of the phosphorylethanolamine in the brain tissue of young animals is only 70 per cent of that of the total acid-soluble fraction, while that of phosphorylcholine is higher, acquiring radioactive phosphate at a rate greater than the majority of other components, but coming into equilibrium with them after 24 hr. As a result of the second observation it was suggested that the choline phosphokinase of nervous tissue must be particularly active *in vivo*. After long exchange periods, however, nothing can be deduced about the relative rates of incorporation of phosphorus-32 into these components of the brain acid-soluble fraction (Fig. 1) and it is likely that all its phosphorus-containing components are in isotopic equilibrium (cf. ref. 11). Miani and Bucciante[7] have presented results indicating that, for animals 6 days old, the incorporation of phosphorus-32 into brain phosphorylethanolamine and phosphorylcholine is maximal 40 hr. after injection, but they do not record the values for the specific radioactivity of the total acid-soluble phosphorus.

It can be seen that several of the phospholipids steadily acquired radioactivity over a time period of seven days (Fig. 2) as was originally observed for phospholipid phosphorus as a whole for both young and adult rats by Changus, Chaikoff and Ruben[12] and afterwards for young rats by Davison and Dobbing[13]. Phosphatidyl serine acquired it at the slowest rate and phosphatidyl choline at a faster rate than that of the ethanolamine-containing phospholipids (cf. ref. 2). Loss of radioactivity from rat brain phospholipids as a whole is apparently very slow[13].

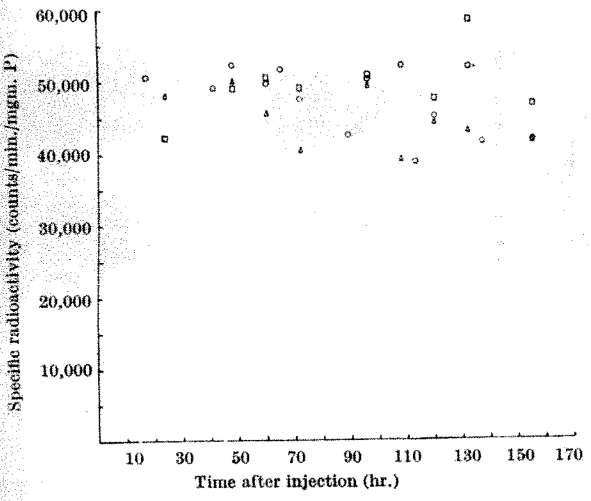

Fig. 1. The specific radioactivity-time scatter diagrams for rat brain acid-soluble phosphorus (○), phosphorylcholine(△) and phosphorylethanolamine (□) after injection of phosphorus-32 labelled orthophosphate when the animals were three weeks old

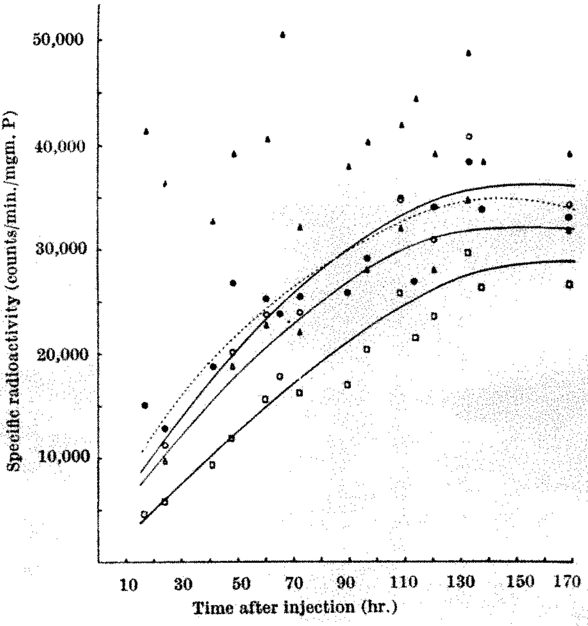

Fig. 2. The specific radioactivity-time curves for rat brain 'diphosphoinositide' (▲), phosphatidic acid (broken line ●), phosphatidyl choline (continuous line ○), phosphatidyl ethanolamine (continuous line △) and phosphatidyl serine (continuous line □)

It was shown previously[3] that 'diphosphoinositide' (more probably a monophosphoinositide[14]) acquired labelled phosphate faster than any other phospholipid and after a short time interval of 3 hr. the specific radioactivity of its phosphorus was about half that of the acid-soluble phosphorus. Phosphorus exchange was apparently proceeding, in fact, at a rate not very different from that of the phosphorylethanolamine in the acid-soluble fraction. The values in Fig. 2 show that after its initial steep rise the specific radioactivity of the inositol phospholipid remained relatively constant; the scatter of the values is due to the fact that the amount of phosphorus estimated in this instance is very small compared with the radioactivity present. Miani and Bucciante[7] have published curves for the long-term incorporation of phosphorus-32 into brain phospholipids of very young rats (4 days old) and of rats of 100 gm. body-weight. Their values for the young animals indicate a sharp peak for the incorporation of phosphorus-32 into 'diphosphoinositide' about 20 hr. after administration of the isotope. The specific radioactivity then rapidly falls away; in the older animals the peak occurs about 60 hr. after administration and is flattened; the specific radioactivity then falls away more slowly. It is difficult to correlate these results with our own observations; for the older animals of Miani and Bucciante, incorporation of label into phosphatidyl choline, phosphatidyl ethanolamine and phosphatidyl serine is the reverse of that found by us for animals three weeks old. For the very young animals in the experiments of the Italian workers, the incorporation in the first 15 hr. following injection would appear similar to that observed by Ansell and Dohmen[2]. Afterwards, the curves follow different courses. It might be pointed out here that in the original paper of Ansell and Dohmen[2] the phosphatidic acid is referred to as an 'unknown phospholipid' yielding glycerophosphate on mild alkaline hydrolysis, because of the doubt then existing about the presence of phosphatidic acid as an *in vivo* component of tissues. Meanwhile, Hokin and Hokin[14] have resolved this problem and have shown that phosphatidic acid is present in brain tissue, though their results suggest that its uptake of phosphorus-32 over a short interval *in vivo* is slower than we have found. Hübscher and Clark have also unequivocally obtained a phosphatidic acid from liver[15].

It is, however, admittedly difficult to make comparisons when animals of different ages are involved. For the rat *in vitro*[16] and for the rat and mouse *in vivo*[1,17] it has been shown that the capacity of the brain to incorporate orthophosphate into phospholipids declines rapidly from birth to early maturity; thereafter the rate decreases much less sharply. Recent experiments have shown that brain sphingomyelin incorporates radioactive phosphate much more rapidly in the myelinating rat brain than in the older brain when myelin is not being so actively formed[18]. It is apparent, in fact, as might be expected, that the rate of replacement of the phosphate moiety of brain phospholipids varies with age and with the phospholipid under consideration.

[1] Dawson, R. M. C., and Richter, D., *Proc. Roy. Soc.*, B, **127**, 252 (1950).

[2] Ansell, G. B., and Dohmen, H., *J. Neurochem.*, **2**, 1 (1957).

[3] Davison, A. N., and Dobbing, J., *Biochem. J.*, **73**, 701 (1959).

[4] Marinetti, G. V., Erbland, J., Albrecht, M., and Stotz, E., *Biochim. Biophys. Acta*, **30**, 543 (1958).

[5] Ansell, G. B., and Dawson, R. M. C., *Biochem. J.*, **50**, 241 (1951).

[6] Ansell, G. B., and Spanner, S., *J. Neurochem.*, **4**, 325 (1959).

[7] Miani, N., and Bucciante, G., *Atti. Soc. Med. Chir. Padova*, **34**, 5 (1956–57).

[8] Dawson, R. M. C., *Biochim. Biophys. Acta*, **14**, 374 (1954).

[9] Ansell, G. B., and Dohmen, H., *J. Neurochem.*, **1**, 150 (1956).

[10] Dziewiatkowski, D., and Bodian, D., *J. Cell. Comp. Physiol.*, **35**, 141 (1950).

[11] Samuels, A. J., Boyarsky, L. L., Gerard, R. M., Libet, B., and Brust, M., *Amer. J. Physiol.*, **164**, 1 (1951).

[12] Changus, G. W., Chaikoff, I. L., and Ruben, N., *J. Biol. Chem.*, **126**, 493 (1938).

[13] Davison, A. N., and Dobbing, J., *Lancet*, 1158 (1958).

[14] Hokin, L. E., and Hokin, M. R., *J. Biol. Chem.*, **233**, 800 (1958).

[15] Hübscher, G., and Clark, B., *Biochem. J.*, **72**, 7*P* (1959).

[16] Fries, B. A., Schachner, H., and Chaikoff, I. L., *J. Biol. Chem.*, **144**, 59 (1942).

[17] Fries, B. A., Changus, G. W., and Chaikoff, I. L., *J. Biol. Chem.*, **132**, 23 (1940).

[18] Ansell, G. B., and Spanner, S., *Biochem. J.*, **73**, 3*P* (1959).

METABOLISM OF POLYPORES WITH PHOSPHORUS-32

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Kar Medical College, Calcutta

IN these studies, our object is to trace the migration of phosphorus-32 from the host-plant to the fungus as well as its migration from the surface of the fruit-body of the fungus to its hymenial layer by direct application on the upper surface of the fruit-body. No such study has been carried out with higher fungi, so far as can be ascertained from published reports. Recently, Grossbard[1] discussed autoradiography in the metabolism of fungi, showing its application in the physiology of some fungi (mostly lower) and pointing out its future usefulness in mycology and plant pathology.

Phosphorus-32 has been one of the most widely used radioisotopes on account of the biological importance of the element, as it is well known that phosphorus enters into the constitution of protoplasm, of phospholipids, phosphoproteides, amino-acids, and neucleoproteides (cellular metabolism); moreover, phosphorus-32 can be easily produced, it has convenient half-life (14·3 days) and it emits energetic β-rays.

We first followed absorption of phosphorus-32 by *Fomes* (*Polyporus*) *lignosus*. *F. lignosus* is usually known as a common root disease-fungus. Its habitat, distribution, morphology and cultural characters have been fully described by one of us[2].

We adopted two concentrations of phosphorus-32 (solution of phosphorus-32 without carrier in neutral solution of sodium phosphate (*p*H 7) prepared by C.E.A. in sterilized vessels) for our study; 5 μc. and 50 μc. per litre; this activity is sufficiently low as not to disturb the metabolism of the fungi, and with this it is convenient to have reasonably accurate radioactivity measurement. Fresh carrot roots were surface-sterilized with calcium hypochlorite (200° chlorometric per kgm.) for 1½ hr., then washed thoroughly with four or five changes of sterilized water at intervals of 20 min. in order to eliminate the toxic matter completely. The outer surface (peripheral tissues) of such roots, which turned white and translucent due to the treatment, was rejected. They were cut aseptically into pieces of 3–4 cm. in length and put (leaf-side up and root-side down) inside sterilized Roux tubes, constricted ends of which contained Gautheret's[3] liquid medium supplemented with phosphorus-32 (sodium phosphate) of the above two doses; each Roux tube contained one piece of the carrot, and the lower end of each piece was kept dipped in the liquid medium for direct absorption of phosphorus-32. A large number of Roux tubes was thus prepared for tissue culture of carrot root, and after an interval of 15 days, when there was good absorption of phosphorus-32 by the carrot according to the Geiger–Müller count (in many cases there was a small growth of new tissue (callus) at the cut surface of carrot pieces), the Roux tubes containing the carrot pieces were exposed to a constant temperature of 60° C. for 20–30 min. After cooling, each carrot piece was inoculated with a very young hyphal culture of *F. lignosus*, the inoculum being approximately equal in all cases. The fungus (a weak parasite) does not grow easily on living mature carrot roots on account of the tissue-resistance, though it kills at once very young tissue cultures; but it has been found that, if the resistance is lowered by exposure to 60° C. for about half an hour, the fungus makes a vigorous growth (Fig. 1) in the course of two or three days. Now, after an interval of 8–10 days, the measurements of activity of the fungus and of carrot pieces were taken; the activity of the fungus growing on carrot with concentration of 50 μc. and 5 μc. per l. was distinct; detailed results are given in Table 1*a*. This is an example of two techniques together: the technique of tissue-culture of carrot and that of radioisotope of phosphorus-32.

We then studied absorption of phosphorus-32 from solid medium with 1 per cent agar in Petri plates in Gautheret's medium, to which two doses of 5 μc.

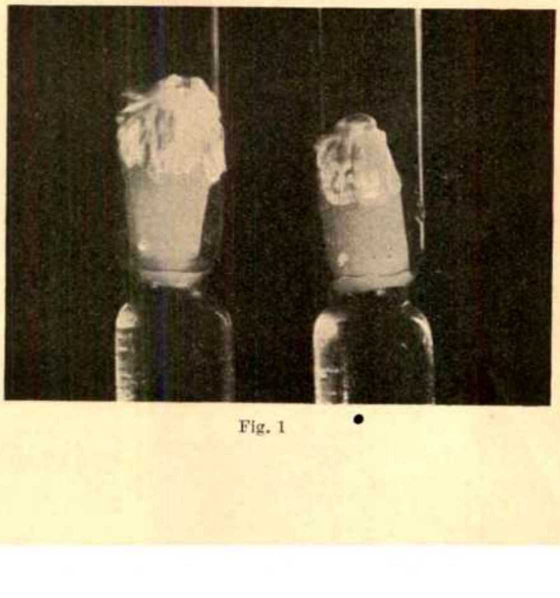

Fig. 1

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Table 1. RADIOACTIVITY OF SAMPLES

(*a*) Activity of fresh tissue.		
Carrot (5 μc./litre)	4·02	counts/min./mgm.
Fungus (*Fomes lignosus*)(50 μc./litre)	270	,,
(5 μc./litre)	389	,,
(*b*) Activity of fresh tissue.		
Agar medium (50 μc./litre)	4·8	,,
Fungus (*F. lignosus*)	69·5	,,
(*c*) Activity of spore-slides of *Ganoderma applanatus* var. *australis*.		
Sporophore *A* (mean of 22 slides)	9·6	counts/min./slide.
Sporophore *B* (mean of 15 slides)	29·2	,,
(*d*) Activity of sporophore-tissues of *G. applanatus* var. *australis*.		
Sporophore *B* context area	24·2	counts/min./cm.^{2}
Sporophore *B* porous area	83·7	,,
Sporophore *C* total area	102	,,

per l. and 50 μc. per l. of phosphorus-32 were added, it being solidified with 1 per cent agar; 10 c.c. of the medium was poured on 9 cm. plates, which were autoclaved, and circular sheets of sterilized 'Cellophane' paper and filter paper equal in diameter to that of the bottom half of a Petri dish were aseptically placed on the surface of the medium, one to each plate. Equal pieces of inocula from the growing edge of a very young plate culture of *F. lignosus* aseptically cut with a cork borer of 8 mm. diameter, were placed on the paper in the plates. After allowing the fungus to grow and absorb phosphorus-32 for 12–20 days, the absorption of the isotope by the fungus at different dates within the interval was measured from the specific activity of the samples according to Geiger–Müller count. The absorption was found significant and that by the bare filter paper was much less than the absorption by the fungal hyphæ; detailed results are given in Table 1*b*.

More interesting results were obtained by application of phosphorus-32 to an intact plant (a *Tilia* tree growing in the compound of the Château de Gif at the Centre National de la Recherche Scientifique) which bore nine small fruit bodies of *Ganoderma applanatus* var. *australis* on the body of the trunk within a length of about 4 ft. above ground-level (Fig. 2 marked with arrows). Scott Russell[4] holds the appearance of radioactivity in shoots of intact plants can provide unequivocal evidence of active transport. Tukey[5] has shown that radioactive minerals entered even through the bark of fruit trees (apple and peach) quickly, and that within 24 hr. of application, radioactivity was detected within the dormant branches, 18–24 in. above and below the points of application, even during winter with temperatures below freezing point.

On July 11, 1958, two square holes (2 cm.^{2}) were drilled in the bark of the trunk of the above-mentioned living tree (*Tilia* sp.) at a height of about 4 ft. from its base, just above and below a sporophore (*A*) of *G. applanatus* var. *australis*, and two cotton pads (2 cm.^{2}) soaked with 2 c.c. of phosphorus-32 (dose being 100 μc. per 2 c.c.), were inserted into the holes, one to each hole; now a number of slides smeared with glycerine were placed below the sporophore on a thick 'Cellophane' platform to catch the falling spores which were being shot out copiously and continuously day and night from the beginning of July, and the whole was very carefully covered with 'Cellophane' paper to keep out external draught or rain. Another cotton pad similarly soaked with phosphorus-32 of the same dose was placed on the top surface of another sporophore (*B*) on the same trunk, and slides (smeared with glycerine) were placed beneath the sporophore, the whole being similarly covered with 'Cellophane' paper. An adjacent sporophore (*C*) just 4 in. below the sporophore (*A*), to which no phosphorus-32 was applied in any way, was kept for observation with slides beneath the sporophore to catch the spore-fall. After an interval of 4 days, slides with copious spore-deposits from the three sporophores *A*, *B* and *C*, as well as fragments of three fruit bodies with porous areas, were taken out for measurement of radioactivity. In each case of the spore-slides and pieces of sporophore the activity was significant, and shot-out spores were distinctly radioactive, while spore-deposit from the same fungus taken two weeks before the application of phosphorus-32 to the trunk showed no radioactivity; detailed results are given in Table 1*c*. The porous areas were more radioactive than the context areas (Table 1*d*). The radioactivity of the sporophore *C*, to which there was no application of phosphorus-32, goes to show that there was active transport of phosphorus-32 in the body of the tree as remarked by Tukey.

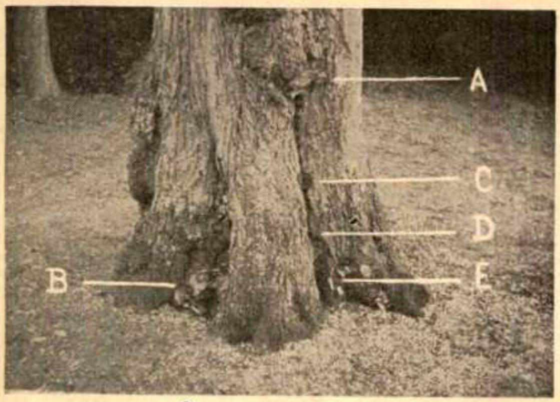

Fig. 2

On October 2, 1958, at 11 a.m., a second higher dose of 1 mc. per 2 c.c. of phosphorus-32 was injected into the top hole only of sporophore *A* (*G. applanatus* var. *australis* growing on the same *Tilia* tree at the Château de Gif), and a 2 per cent agar opened (cover taken out) plate and 10 aluminium cupules were aseptically placed underneath the sporophore at 3 p.m. Geiger–Müller counting after 20 hr. showed that spores were positively radioactive of the order of the background. Before this second injection, spore-fall of *G. applanatus* var. *australis* on slides smeared with glycerine was taken for 24 hr. during September 14–15, 1958; such spores, when tested, were found to be not radioactive. This occurred after an interval of two months and four days from the first injection. Spore-fall from the sporophore *A*, which was radioactive, occurred only once, on October 3; and it entirely stopped thereafter, that is, one month earlier than the normal time, whereas the spore-fall from the sporophore *B* (which was not radioactive) on the same tree continued up to November 6. Germination of spores which were not

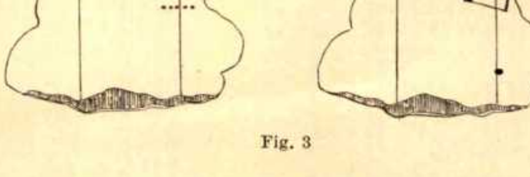

Fig. 3

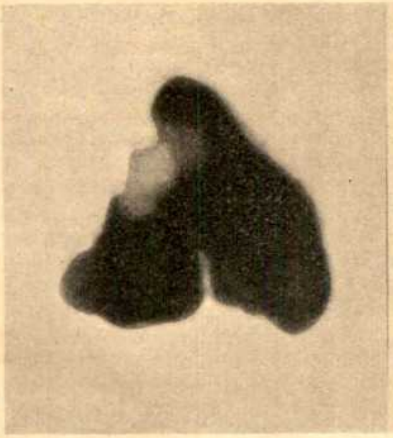

Fig. 4. *D*1. 10 min. exposure

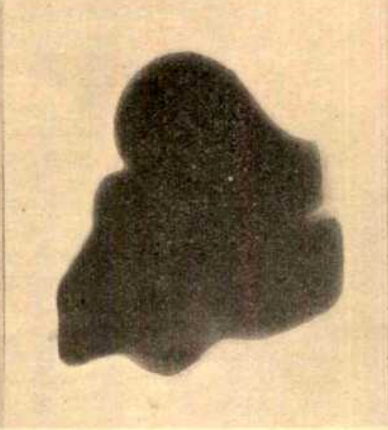

Fig. 5. *D*2. 10 min. exposure

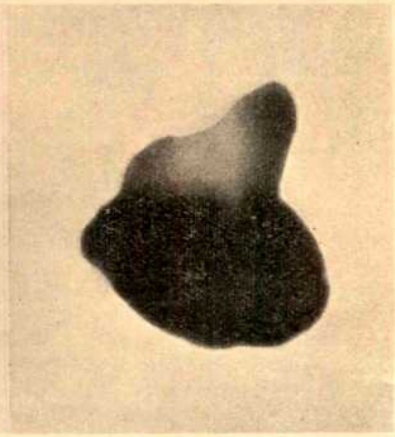

Fig. 6. *E*2. 10 min. exposure

radioactive, in malt-agar tubes, was quite easy; they germinated in the course of 10 days, whereas germination of radioactive spores took place only once, after an interval of 20 days, and in one malt agar tube only out of ten such tubes; but cultures of these radioactive spores in malt agar plates showed no trace of radioactivity. Moreover, from examination of colour change of the hymenial surface of the sporophore *A* from a whitish to a brownish colour after October 3, it appears that it had then stopped throwing out spores (Bose[6]). It was further noted that subcultures of radioactive spores in malt agar plates in the dark at 24° C. (incubator) grew perceptibly more slowly than those of normal non-radioactive spores of *G. applanatus* var. *australis*; the diameter of 10 days growth of the mycelial mat in malt agar plates was only 6 cm. in the case of radioactive spore-hyphæ, whereas in the case of non-radioactive spore-hyphæ the diameter of growth under identical conditions was 8·5 cm. on the average. But detailed examination of the growth of the two mycelial mats under the microscope showed no difference whatsoever in characters of the hyphæ; thus the noted difference in rate of growth may be purely cytoplasmic, as indicated by Jinks[7]. Consequently, it may be held that radioactive spores of *G. applanatus* var. *australis* are susceptible to radiation.

Microscopical examination of the surface of the hymen of normal sporophores showed that, by November 1958, they were past the sporing stage, basidia being almost entirely replaced by tramal hyphæ—elongations which rapidly fill up the pores—and *Ganoderma* spores are no longer found in sections of sporophores.

On December 12, two sporophores (*D* and *E*) of *G. applanatus* var. *australis* growing on the same *Tilia* tree at Gif Château were treated with phosphorus-32, the dose being 1 mc./c.c. in each case. In the case of sporophore *D* the very tough upper surface was scratched to a depth of 1 cm., and a cotton pad soaked with 1 c.c. of phosphorus-32 was placed directly on the scratched surface, while in the case of sporophore *E*, the upper surface was injected with a tuberculin syringe, 1 cm. deep, 1 c.c. of phosphorus-32 being divided into four injections of equal quantity of the liquid. Both the sporophores, *D* and *E*, were very carefully covered with 'Cellophane' and brown paper for protection against the prevailing light showers of rain and occasional gusts of wind. After an interval of four and a half days, each of the two sporophores (*D* and *E*) was cut longitudinally at two spots—one corresponding to the treated areas (*D*1 and *E*1), and the other, distant areas (*D*2 and *E*2); so, they were marked respectively as *D*1 and *D*2 and *E*1 and *E*2 (Fig. 3).

These four pieces were then autoradiographed at Saclay, Service de Biologie du Centre d'Etudes nucleaires, following closely the method of Pellerin[8]. The injection method was not quite satisfactory on account of suspected leakage from the needle-end pressing against the very tough surface of the sporophore, but in the distant area, *E*2 (Fig. 6), there was a clear indication of accumulation of phosphorus-32 towards the hymenial surface. The specimen-pieces were highly radioactive according to Geiger–Müller count—*D*1 giving 49,000 counts per min. per gm., and *D*2 giving 45,000 counts/min./gm.

An exposure of 10 min. to X-ray film in the dark with liquid nitrogen at – 200° C. was sufficient to bring out distinct images (Figs. 4, 5 and 6). It can be seen that heavy accumulation of phosphorus-32 takes place towards the hymenial (porous) surface. There is thus clear evidence of ready transport of phosphorus-32 to the most active area, full of living cells. Control (a longitudinal section of a normal sporophore) under identical conditions showed a perfectly blank area.

One of us (S. R. B.) is grateful to the Centre National de la Recherche Scientifique, Paris, for a research grant for this work and to the authorities of the Institut du Radium (Laboratoire Curie) for facilities for this work, and we thank M. D. Basu working at Saclay (C.E.A.) for help with a portion of this work with tree injection and with part of the Geiger–Müller counting. For the mycological work one of us (S. R. B.) is indebted to Prof. R. Heim for facilities accorded in the Laboratoire de Cryptogamie, Muséum National d'Histoire Naturelle, Paris.

[1] Grossbard, E., *Nature*, **182**, 854 (1958).
[2] Bose, S. R., *Trans. Brit. Mycol. Soc.*, **40**, 456 (1957).
[3] Gautheret, R. J., "Manuel technique de Cultures des Tissus Vegetaux" (Masson et Cie, Paris, 1942).
[4] Scott Russell, R., "Progress in Nuclear Energy", Series VI, Biol. Sciences (Pergamon Press, **1**, 87, 1956).
[5] Tukey, H. B., and Wittwer, S. H., "Progress in Nuclear Energy", Series VI, Biol. Sciences (Pergamon Press, **1**, 106, 1956).
[6] Bose, S. R., *J. Ind. Bot. Soc.*, **23**, 155 (1944).
[7] Jinks, J. L., *C.R. Lab. Carlsberg., Ser. Physiol.*, **26**, 183 (1956).
[8] Pellerin, P., *C.R. Acad. Sci., Paris*, **244**, 1555 (1957).

LETTERS TO THE EDITORS

PHYSICS

Film Transfer in He II

During the course of experiments on film transfer in very narrow beakers and in vessels with very narrow apertures, phenomena have appeared which are difficult to reconcile with the existing interpretation of the nature of film transfer and of critical velocity[1].

It has been found, especially at temperatures in the neighbourhood of 1·3° K. where most of the measurements have been made, that the rate of transfer out of a narrow cylindrical beaker of, say, 1·5 mm. diameter is dependent on the history and condition of the liquid in the beaker and is no longer the characteristic rate of transfer per cm. width of surface at the temperature in question. Eselson and Laserev[2] observed a similar effect some years ago, but were unable then to offer an explanation. The present results are in agreement with their work and extend it in several ways. Recently, Snyder and Donnelly[3], describing film transfer from rotating vessels, also mention one or two instances of a similar effect.

For the experiments reported here a cylindrical beaker was used, 5 cm. long and of inner diameter 1·8 mm. and outer diameter 3·6 mm. The rim of the vessel was ground concave conically so as to give clearly defined outer and inner perimeters. A typical experiment consisted first of filling the beaker—either by plunging it beneath the bath or by allowing it to fill by film transfer—and then of observing a sequence of emptyings and fillings by film transfer alone, over a range of 4–15 mm. below the rim of the beaker. Table 1 shows the results of such an experiment. Sequence *A* was initiated by plunging the beaker and then withdrawing it; sequence *B* was initiated by emptying the beaker completely after *A* and then filling solely by the film; and finally, sequence *C* was initiated by a further plunge-fill of the beaker.

From this and similar experiments with other vessels the following general features were observed: (*a*) following a plunge-fill, the rate of emptying was higher than it was after a film-fill, the maximum observed difference between the two rates for the same beaker being nearly 70 per cent; (*b*) rates of filling were always lower than rates of emptying, the difference being to some extent dependent on the wall thickness; (*c*) for the same vessel, rates of emptying after successive plunge-fills differed from each other, sometimes by as much as 15 per cent; (*d*) a pause of 20 min. between a plunge-fill and subsequent emptying had no observable effect of reducing the fast rate of emptying.

Table 1

Sequence	Transfer-rate on emptying (cm.^{2} sec.$^{-1}$ × 10^5)	Transfer-rate on filling (cm.^{2} sec.$^{-1}$ × 10^5)
A (Plunge fill)		
Emptying	13·2	
Filling		6·87
Emptying	9·02	
Filling		6·82
Emptying	9·17	
B (Film fill)		
Filling		6·90
Emptying	8·57	
Filling		6·70
Emptying	9·47	
C (Plunge fill)		
Emptying	14·2	
Filling		6·42

It was inferred that the fast rate of emptying by the film following a plunge-fill drained off the excess of something which had been introduced by the plunge, and that this excess might be turbulence. This inference was tested in a separate experiment in which a small solenoid-activated iron wire stirrer was put into a beaker. It was observed that continuous stirring during emptying increased the rate of emptying by 10 per cent over the unstirred rate, while continuous stirring during filling reduced the normal filling rate by about 10 per cent. Although the reduction in filling rate was remarkable, the increase in emptying rate caused by stirring the beaker was not as great as the increase obtained by a plunge-fill, which might indicate that stirring is not an efficient way of making turbulence in so nearly an ideal fluid.

It has been assumed that there exists a 'critical' rate of film transfer, although its precise value has been a matter of some disagreement. Hence, knowing the thickness of the film one can determine a 'critical' velocity. From earlier experiments it has been inferred that when the velocity is less than 'critical' the liquid in the film moves without friction, but that at or a very little above the 'critical' velocity the friction becomes great and the energy is dissipated in the creation of excitations.

It is difficult to reconcile the notion of 'critical' velocity with the experiments that are reported here, and we may therefore be forced to abandon it. If so, it could be replaced by one in which the movement of the film is intimately associated with turbulence. Suppose, for example, that the film moves, as it were, on line vortex roller bearings, and that if no such vortices are present in the film then no motion of the film occurs. If this is so, then one might expect that the more vorticity there is available at the source of the film the more liquid will be transferred by the film, whereas if vorticity is scarce at the source then the rate of transfer will be lowered. Excess of turbulence at the downstream end of the film also appears to slow the transfer, which might indicate either that turbulence is flowing both ways in any film so that the transfer is a net transfer, or that the specific quantity of turbulence in a vessel has an appreciable chemical potential.

The picture, although it is obviously very crude, does not appear to be in conflict with earlier experiments on film transfer, and it may also have relevance for the concept of critical velocities in the bulk liquid. A so-called 'critical' velocity might cease to be the upper limit of velocity before the onset of friction, and become a velocity which depends on the energy required to produce the amount of vorticity involved in the flow.

This kind of vorticity or turbulence is very long-lived; in the example quoted earlier it appeared to remain more or less intact for at least 20 min. in a volume of about 0·1 cm.[3]. The conversion of such vorticity into rotons, which was envisaged by Feynman[4], seems therefore to be a very slow process indeed.

In the particular case of film transfer one is involved in discovering the particular properties of line vortices running along close to a wall, and also whether in the film which covers the surface of a tube the vortices form closed rings or a continuous helix with one end fixed in the bath and the other in the beaker.

These results, together with others made with vessels with very narrow necks or with matt surfaces, will be more fully discussed in a later paper.

In conclusion, I wish to record my thanks to Dr. C. G. Kuper for much fruitful collaboration, and also to Dr. D. V. Osborne and Dr. D. Bohm for stimulating discussion.

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[1] See, for example, Atkins, K. R., "Liquid Helium" (Cambridge University Press, 1959).

[2] Eselson, B. N., and Laserev, B. G., *J. Exp. Theor. Phys.* (U.S.S.R.), **23**, 552 (1952).

[3] Snyder, H. A., and Donnelly, R. J., *Phys. Fluids*, **2**, 408 (1959).

[4] Feynman, R. P., *Prog. Low Temp. Phys.*, **1**, 17 (1955).

Mechanism of Superfluid Flow

Theories of superfluidity usually assume the possibility of pure potential flow, provided that the flow velocity is less than some critical value (at which the creation of excitations first becomes possible)[1,2]. If we consider the flow of helium (at $T = 0°$ K.), for example through a tube or along a Rollin film, it is implied that the boundary layer is slipping over the solid wall. However, the first few layers of helium atoms would not be expected to take part in this motion, as they are firmly bound by the van der Waals' attractions of the wall. The picture therefore has to be revised slightly to one in which there is a velocity discontinuity (that is, a vortex sheet) close to, but not coincident with, the solid wall. But a continuous vortex sheet implies a rather drastic change in the character of the wave function, over a distance of the order of one interatomic distance a, and hence implies a large energy per unit area, of the order of $\hbar^2/ma^4$ (where m is the mass of an atom).

A more realistic model for helium, flowing at $T = 0°$ K., would seem to be one in which the vortex sheet is replaced by some sort of array of Onsager–Feynman quantized vortex lines. For example, a Kármán row of unit quantized vortices[3] will be able to reproduce the macroscopic features of the 'vortex sheet' model for flow parallel to a plane wall; the vortices will themselves take part in the motion. The liquid in contact with the wall should have zero average velocity and reasonably small actual instantaneous velocity.

Let us consider a row of vortex lines of strength $\varkappa = 2\pi\hbar/m$, perpendicular to the plane of the paper, cutting it at the points (na, b), and let the solid surface be the plane $y = 0$ (Fig. 1). Then, if the vortices are at rest, the x and y components of the velocity are given by the real and imaginary parts respectively of[3]:

$$-i\pi\varkappa a^{-1}\cot\{\pi a^{-1}(x + i(y - b))\} \qquad (1)$$

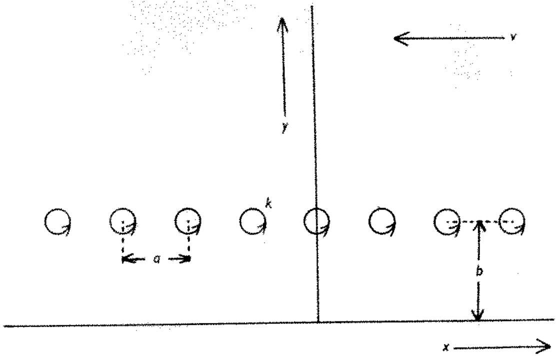

Fig. 1

If $a \ll b$, this pattern approximates to a vortex sheet, and the oscillatory part of the velocity is very small except very near the vortex row; indeed, even if a is fairly comparable to b, provided only $\exp(-b/a) \ll 1$ holds, the periodic fluctuations of velocity at the wall remain small compared to the zero-point velocities of helium atoms. The mean velocity on a plane parallel to the wall is:

$$-\pi\varkappa a^{-1}\tanh\{\pi(y - b)/a\} \qquad (2)$$

and is in the x-direction. The boundary layer can be brought to rest, on the average, by superimposing a velocity:

$$-\pi\varkappa a^{-1}\tanh(\pi b/a) \qquad (3)$$

on the fluid (and in particular on the vortex lines). The magnitude of the macroscopic velocity of flow is then:

$$V = 2\pi\varkappa a = 4\pi^2\hbar/ma \qquad (4)$$

provided $b \gg a$, and the depth of liquid $\gg b$.

In a Rollin film of thickness t, if $a \gg t$, flow by this mechanism cannot proceed—there are not enough vortices to serve as 'bearings' (Allen, J. F., previous communication). Hence a film cannot flow steadily unless $a \lesssim t$, when:

$$V \gtrsim 4\pi^2\hbar/mt \qquad (5)$$

Thus, instead of a 'critical' velocity, h/mt plays the part of a minimum flow velocity[4]. Any higher velocity is possible, but in general energy will have to be supplied to replace the vortex lines carried away by the flow. For the occurrence of lower velocities, a more complicated configuration of vortices will be needed, which will probably lead to increased dissipation. Some instability of flow might also be expected near the velocity (5).

Allen has suggested that his experimental results should be interpreted as showing that a Rollin film flows by 'rolling' on vortices. The microscopic arguments advanced here lead to just this picture of the flow. Older experiments, which appear to support the idea of a critical velocity, require re-interpretation. The present suggestion is that there is a velocity of minimum energy dissipation—the velocity which is normally observed in steady flow. It should be noted that a persistent current around a torus remains possible, since vortex rings can just roll round the torus.

The actual geometrical array of vortices is likely to be more complicated than I have envisaged here. The energy of interaction between two vortex lines is quite comparable to Feynman's[2] self-energy of one line, so that quantitative calculations become difficult. The mutual repulsions between like parallel vortices will probably tend to give a fairly uniform lattice of lines, rather than just a row.

I am greatly indebted to Prof. J. F. Allen for many discussions during the progress of his experiments and wish also to thank Dr. D. Bohm and Dr. O. Penrose for stimulating discussions.

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[1] Landau, L. D., *J. Phys.* (U.S.S.R.), **5**, 71 (1941); **11**, 91 (1947). Feynman, R. P., *Phys. Rev.*, **94**, 262 (1954).

[2] Feynman, R. P., "Low Temperature Physics", **1**, 17 (North Holland Pub. Co., Amsterdam, 1955). Onsager, L., *Nuovo Cim.* (ix), **6**, Supp., 249 (1949).

[3] See, for example, Milne-Thomson, L. M., "Theoretical Hydrodynamics", 340 (Macmillan, London, 1948).

[4] Contrast with the conventional interpretations; see, for example, Atkins, K. R., "Liquid Helium", 90 (Cambridge University Press. 1959).

Measurements of the Frequency of an Ammonia Maser in England and Australia

THIS communication describes preliminary measurements of the oscillation frequency of the Signals Research and Development Establishment Maser No. 5 at the National Physical Laboratory, Teddington (by the Standards Division), and afterwards at the Research Laboratories of the Australian Post Office in Melbourne, Australia. Maser No. 5 is a rack-mounted experimental maser with continuous pumping and employs unpurified commercial anhydrous ammonia. An eight-pole state separator is used together with a silver-plated brass microwave cavity operating in the TM_{010} mode. The cavity is thermally tuned to the (3,3) transition. A more detailed description of this maser may be found elsewhere[1].

The maser was operated at the National Physical Laboratory under the following conditions: Ammonia gun pressure 1·0 mm. mercury (corresponding to a total flow into the system of 3×10^{17} mole./s.) and 18 kV. d.c. between adjacent poles of the state separator. The resonant frequency of the cavity was adjusted thermally to a value such that the maser oscillation frequency changed less than 2 parts in 10^{10} when the gun pressure was changed from 0·5–1·0 mm. mercury. This was one of several possible methods of establishing a resettable oscillation frequency. Other methods give different oscillation frequencies[1,2], none of which necessarily corresponds to the emission line frequency. The oscillation frequency was measured in terms of a local 5 Mc./s. quartz standard which in turn was calibrated against the cæsium resonator[3]. Results are shown in Table 1 (sets 1 and 2), the absolute accuracy of the National Physical Laboratory measurements being ± 2 parts in 10^{10}. After completion of set 1 the state separator and ammonia gun assembly were removed and the wave-guide connexion to the cavity uncoupled. This was an attempt to simulate the dismantling required prior to shipment of the maser by sea to Melbourne. After re-assembly the maser frequency was remeasured with the results shown as set 2. The downward shift in frequency of 1·3 parts in 10^9 is significant but time did not permit investigation of the reason.

The maser was then dismantled, shipped to Melbourne and installed at the Australian Post Office Research Laboratories. Certain improvements were made to the thermostat controlling the microwave cavity temperature and the cavity frequency again adjusted so that the maser oscillation frequency was independent of gun pressure changes between 0·5 and 1·0 mm. mercury. As before, the gun pressure was maintained at 1·0 mm. mercury and the separator voltage at 18 kV. d.c., while measurements of oscillation frequency were made in terms of a local 100 kc./s. quartz standard. The results are shown as sets 3–8, the spread of measurements within a set (col. 6) being slightly inferior to that obtained at the National Physical Laboratory. The frequency of the local quartz standard was measured by comparing it with the G.B.R. transmission from Rugby using a beat frequency method. Because of the poor signal-to-noise ratio of the received signal it was necessary to average the beats between G.B.R. and the local standard over a period of 22 hr. each day to obtain an accuracy of ± 4 parts in 10^{10}. However, an additional but unknown error of perhaps several parts in 10^9 must also be added due to propagation variations over the path Rugby–Melbourne. After applying the National Physical Laboratory corrections to the oscillator controlling G.B.R., the mean maser frequencies shown in sets 3–8 are obtained. The grand average of all the Melbourne results is 2·2 parts in 10^9 higher than the average of the National Physical Laboratory results. As stated above, this frequency difference is comparable to the propagation error which must be attached to the Melbourne mean frequencies given in Table 1.

Some evidence has been obtained to show that the frequency of oscillation is dependent on the ambient temperature to the extent of −1 part in 10^{10}/deg. C. rise. This is almost certainly due to inadequacy of the thermostat controlling the cavity temperature. The effect can be seen in sets 6, 7 and 8. The ambient temperature was rising throughout November 11, 1959, and was higher during measurements on November 12, 1959. Further work is being carried out both at the Signals Research and Development Establishment and the Australian Post Office Research Laboratories, Melbourne, towards discovering and controlling the residual factors governing the maser oscillation frequency. For comparison pur-

Table 1. SUMMARY OF FREQUENCY MEASUREMENTS ON SIGNALS RESEARCH AND DEVELOPMENT ESTABLISHMENT MASER NO. 5
Assumed cæsium frequency = 9 162 631 770 c./s.

Measurement set No.	Maser location	Date	Mean maser frequency (c./s.)	No. of readings	Standard deviation (parts in 10^{10})	Deviation of mean frequency from set 2 (parts in 10^9)
1	Teddington	14. 5.59	23 870 129 145	46	2·6	+ 1·3
2	,,	20. 5.59	,, ,, ,, 115	82	1·9	0
3	Melbourne	6.11.59	,, ,, ,, 197	5	3·2	+ 3·4
4	,,	9.11.59	,, ,, ,, 178	40	3·0	+ 2·6
5	,,	10.11.59	,, ,, ,, 196	41	2·8	+ 3·4
6	,,	11.11.59	,, ,, ,, 184	41	3·6	+ 2·9
7	,,	11.11.59	,, ,, ,, 167	47	4·1	+ 2·2
8	,,	12.11.59	,, ,, ,, 149	20	6·2	+ 1·4
—	Neuchâtel	—	,, ,, 128 870	—	—	−10·3

poses we have appended to Table 1 the published frequency[4] for an ammonia maser employed at Laboratoire Suisse de Recherches Horlogères, Neuchâtel, using another re-setting criterion. The frequency of this maser is considerably lower than that of Maser No. 5 and illustrates the fact that it is possible to operate ammonia masers with considerably different but yet highly stable oscillation frequencies.

We wish to thank the Director of the National Physical Laboratory for making available the results of the maser/cæsium comparisons obtained at Teddington. This communication is published with the permission of the Chief Scientist, Ministry of Aviation, and the Engineer-in-Chief, Post Master General's Department, Commonwealth of Australia.

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[1] Mitchell, A. M. J., Roots, K. G., and Phillips, G., "Electronic Technology" (to be published).

[2] Barnes, F. S., *Proc. Inst. Rad. Eng.*, **47**, 2085 (1959).

[3] Essen, L., and Parry, J. V. L., *Trans. Roy. Soc.*, **250**, 45 (1957).

[4] Observatoire de Neuchâtel, Suisse, Circulaire No. 2 (May 1959).

Monocular 'Rivalry' between Stabilized and Unstabilized Retinal Images

IT is well known that images stabilized against retinal tremor tend to fade from vision[1,2], and it has been suggested that this indicates some form of fatigue or adaptation in the retinal receptors, which is normally relieved by small irregular displacements of the image. When using a simple stabilizing device[3] for the study of complementary patterns[4], I have observed a related effect which seems to require further, if not different, explanation.

The apparatus[3] comprises a close-fitting contact lens with an optical power of 60 dioptres, supporting a light 'Dural' tube on which objects under examination are mounted at the focal distance of about 1½ cm. The lens incorporates an artificial pupil 4 mm. in diameter. When the image of a sharply black-and-white object such as my 'ray' figure[4] is thus stabilized on the retina, fading is much less pronounced and permanent than with more delicate and irregular gradations of stimulus intensity[2].

The observation in question is as follows. When a thin object, such as a wire the diameter of which subtends perhaps 10′, is viewed in silhouette, it often remains visible for some time. If, however, an object giving an unstabilized image, such as the finger-tip, is now brought up just behind the wire to the focal zone of the stabilizing lens, a striking change takes place. The stabilized wire disappears as a whole, and the finger-tip appears completely visible, as if nothing were between it and the eye. The impression is similar to that given by binocular rivalry between disparate images in left and right eyes. When the finger is removed, the wire reappears.

With stabilized objects of increasing width, it is necessary (as might be expected) to scan the unstabilized background with an increasing amplitude if the objects are to disappear from view, and a limit is reached (dependent partly on the structure of the background) beyond which total disappearance does not occur.

It may well be suspected that the reluctance of the wire (and indeed of the 'ray' figure) to disappear permanently when stabilized is due in part to imperfect stabilization. This I leave an open question, though it may be remarked that with apparatus which presents the stabilized image within an unstabilized frame[1,2] it would now seem unsafe to use extinction of the image as a criterion of stability. What seems significant is that one can cause the disappearance—sudden and complete—of a thin stabilized object merely by presenting it against an unstabilized background. Together with the inhibition of extinction by regularity and boldness in the stabilized pattern this raises doubts whether changes in retinal receptors alone are responsible for the normal fading. On a retinal theory one might have expected that the addition of an unstabilized background would revive activity in neighbouring receptors adapted to a stable image by disturbing them, as flickering light is found to do[5].

It is difficult to see how it could predict the opposite effect actually observed, particularly the simultaneous and sudden disappearance of all parts of the wire. For this the idea of fatigue or adaptation among primitive gestalt-forming mechanisms would seem worthy of exploration.

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[1] Ditchburn, R. W., and Ginsborg, B. L., *Nature*, **170**, 36 (1952).

[2] Riggs, L. A., Ratcliff, F., Cornsweet, J. C., and Cornsweet, T. N., *J. Opt. Soc. Amer.*, **43**, 495 (1953).

[3] MacKay, D. M., *Nature*, **180**, 1145 (1957).

[4] MacKay, D. M., *Nature*, **180**, 849 (1957).

[5] Ditchburn, R. W., and Fender, D. H., *Opt. Act.*, **3**, 128 (1955).

GEOPHYSICS and GEOLOGY

Palæomagnetism in Antarctica

WE have recently measured the direction of magnetization of two dolerite sheets and ten basic dykes in the Wright Valley (lat. 77·5° S., long. 161·5° E.) of South Victoria Land. The dykes intrude the basement granite in the area but do not cut the overlying Beacon Sandstone. The dolerite sheets are intruded into the basement and in some places cut the Beacon Sandstone. They post-date the dykes and the sandstone. The directions and relevant statistics are given in Table 1.

There is good evidence that the directions of magnetization are stable. The divergences of the mean directions of both sheets and dykes from the present Earth's field and the dipole field exceed the errors ($P = 0{\cdot}05$) by a factor of 3 or more. The coercivity of saturation isothermal remanence is high, varying from 600 to 1,200 œrsteds. Treatment in alternating magnetic fields of up to 370 œrsteds (peak) produced only small changes in the directions of magnetization. In some cases the directions remained little changed in alternating fields up to 750 œrsteds. These changes vary between 2° and 5° and are of the same order as the experimental error.

Other pole results from Antarctica have recently been reported by Nagata and Shimizu[1] from the Pre-Cambrian gneiss of Ongul Island (namely, lat. 3° N.,

Table 1. MEAN DIRECTIONS OF MAGNETIZATION AND POLE POSITIONS

Geological unit	N	n	s	Mean direction of magnetization				Pole position				Divergences	
				D	I	R	α	Latitude	Longitude	$\delta\psi$	$\delta\chi$	$\triangle F$	$\triangle P$
Dolerite sills	8	11	20	262	−70	7·889	7	51° S.	132° W.	10	12	23	21
Basic dykes in grey granite	10	14	31	237	−56	9·832	7	29° S.	149° W.	7	10	32	38

N denotes the number of sites, n is the number of samples collected and s is the number of specimens cut from these samples. D and I are the declination and inclination of the resultant direction of magnetization computed by giving each site unit weight. R is the length of the resultant, α is the error of the mean at $P = 0{\cdot}05$ (Fisher, ref. 4), and $\triangle F$ and $\triangle P$ are the divergences of the mean direction from the present Earth's field (155°, −83°) and from the dipole field (0°, −84°) respectively. The pole positions in the southern hemisphere are specified by latitude and longitude and the errors $\delta\psi$ and $\delta\chi$ are in the direction of and at right-angles to the co-latitude, respectively.

long. 107° W.), by Turnbull[2] from the Cainozoic volcanics of Cape Hallett (lat. 81° S., long. 94° E.), dolerite sills of the Ferrar Glacier (lat. 58° S., long. 142° W.) and from the Beacon Sandstone (lat. 53° S., long. 151° W.), and by Blundell and Stephenson[3] from some dolerites from Theron Mountains and Whichaway Nunataks (lat. 54° S., long. 136° W.). These poles, together with those calculated from our results, are plotted in Fig. 1. The results from the dolerites are in good agreement. Turnbull is of the opinion that the Beacon Sandstone he sampled may have been reheated by the dolerite, which thus accounts for the agreement of the result from the Sandstone with the dolerites.

The precise geological age of these rock formations is not known. The basement dykes could be Palæozoic, the Beacon Sandstones are probably for the most part Upper Palæozoic or Early Mesozoic, and the dolerites are very likely to be Mesozoic in age. However, except for 4C, about the age of which little can be said, the relative age sequence is almost certainly that given in Fig. 1. The following general points may therefore be made.

(1) The divergence of the palæomagnetic pole from the present geographical pole increases with geological age. In geological recent times it is in the vicinity of the present pole and in the Pre-Cambrian it is near the present-day equator (see ref. 1 for discussion of the latter point).

(2) The poles have characteristic longitudes between 100° and 150° W. In these two features the results are similar to the pole paths obtained from Europe, North America, India and Australia.

(3) However, the longitudes differ very greatly (in some cases by 100° or more) from the longitudes of the poles obtained from these four regions. This result emphasizes for yet another continent the divergence that occurs between results from different continents for periods prior to the Tertiary.

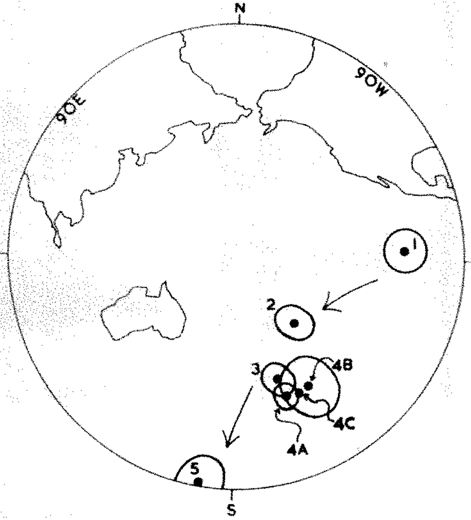

Fig. 1. Pole positions from Antarctica. The pole positions are numbered as follows: 1, Ongul Island gneiss; 2, basement dykes; 3, Beacon Sandstone; 4A, dolerite sills, Ferrar Glacier; 4B, dolerite sheets of Wright Valley; 4C, Theron and Whichaway dolerites; 5, Hallett Cove volcanics. The projection is equatorial stereographic

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[1] Nagata, T., and Shimizu, Y., *Nature*, **184**, 1472 (1959).
[2] Turnbull, G., *Arctic*, **12**, 151 (1959).
[3] Blundell, D. J., and Stephenson, P. J., *Nature*, **184**, 1860 (1959).
[4] Fisher, R. A., *Proc. Roy. Soc.*, A, **217**, 295 (1953).

Infra-Red Determination of the Kaolin Group Minerals

BY infra-red absorption analysis, it is possible to make a semi-quantitative estimation of the kaolin minerals. In contrast to X-ray diffraction, the infra-red spectrum of halloysite is as clearly defined as those of kaolinite and dickite, and the content of these three minerals in mixtures can be determined by a ratio between the depths of their hydroxyl absorptions. The peaks at 2·70μ (A) and 2·75μ (B) are common to all three minerals, and a peak depth ratio A/B of 1·2–1·5 is characteristic of kaolinite, with a ratio A/B of 0·8–0·9 characterizing halloysite. Values of 0·6–0·8 are typical of dickite. If the depth of the band due to physically adsorbed water at 2·9μ (C) is plotted as the abscissa with A/B as the ordinate, the three minerals fall into separate areas.

Examination of the spectra due to Si–O and Al–Si–O between 8·9 and 10·1μ show a diagnostic pair of doublets. The doublet at 10·7μ (D) and 11·0μ (E) is typical of the Al–O–OH bonding of kaolinite and dickite, although only the single peak at 11·0μ appears for halloysite. An inspection of the A/B ratio and the presence, or absence, of absorption (D) enables a unique distinction to be made between the three minerals. The A/B ratio remains constant during dehydration over several hours up to 450° C.

The absorption at 2·70μ (A) is found only in the kaolin group and in such simple structures as brucite. It is due to oxygen–hydrogen stretching frequencies associated with the hydroxyl ions, the oxygen and hydrogen atoms being in two symmetrical layers in

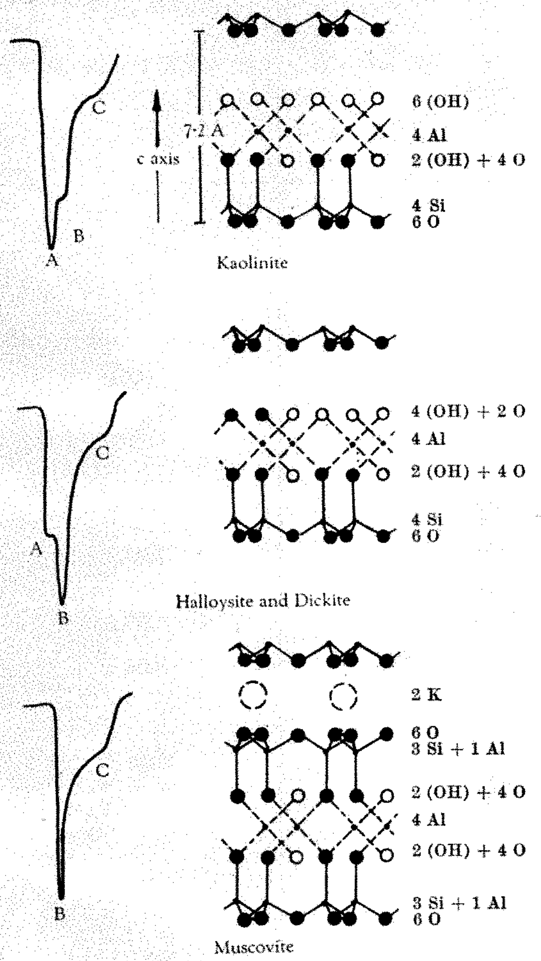

Fig. 1. Suggested oxygen–hydroxyl configuration for halloysite and dickite as compared with kaolinite and muscovite. Hydroxyl portions of their infra-red curves appear to the left of each structure

the brucite crystal. The 2·75μ (*B*) absorption is common to all micas and three-layer minerals, which differ from kaolinite in that their upper layer of oxygen–hydroxyls is only one-third hydroxyl, two-thirds oxygen.

A modification of the general kaolinite structure is proposed to explain the similarity in *A*/*B* ratio in halloysite and dickite as contrasted to that of kaolinite. The frequency of hydroxyls in the upper oxygen–hydroxyl level in the kaolin structure must vary between 3/3 (as in kaolinite) and 1/3 (as in the micas, when both upper and lower levels of the octahedral layer become symmetrical) (Fig. 1). Micas thus have a single (*B*) hydroxyl absorption. Kaolinite itself shows a full upper level of (*A*) type and full lower level of (*B*) type. Halloysite and dickite must lie at an intermediate value where 2/3 of the upper level are hydroxyls. This would decrease the strength of the (*A*) type absorption and increase the strength of the (*B*) type absorption, lowering the *A*/*B* ratio.

Aluminium–silicon substitution (*Y* number) in the basal, tetrahedrally co-ordinated levels of kaolinite, as proposed by Bates[1], should be very easy to determine in the same manner as used in the chlorite and other mica studies[2].

A more complete study on this subject will be presented at the forthcoming Pittsburgh Conference on Analytical Chemistry and Applied Spectroscopy in March 1960.

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[1] Bates, T., and Comer, J. J., Clays and Clay Minerals, Proc. Sixth Nat. Clay Conf., Nat. Acad. Sci.–Nat. Res. Council (1958).

[2] Tuddenham, W. M., and Lyon, R. J. P., *Anal. Chem.*, **31**, 377 (1959).

Sea-Level Fluctuations during the Past Four Thousand Years

Study of constructional details of a chenier plain along the coast of the Firth of Thames, New Zealand, together with radiocarbon dating of seven shell samples, has enabled the production of Fig. 1. Criteria used in interpreting the past sea-levels were (*a*) storm ridges, (*b*) high-spring-tide wash benches and (*c*) tidal stream flats. Often all three criteria could be used to determine one sea-level. Periods of sea-level highs correlate well with periods of transgression recorded along the European coast[1-3].

Tectonic movement, compaction and other causes could have led to misleading results; but their effects are considered small. Thus neglecting the minor fluctuations shown in Fig. 1, sea-level fell 7 ft. from 2000 B.C. to about the beginning of the Christian era and has remained relatively stable since. The present rise of sea-level is locally 8–9 in. per century and may well be only another minor fluctuation in an otherwise stable sea.

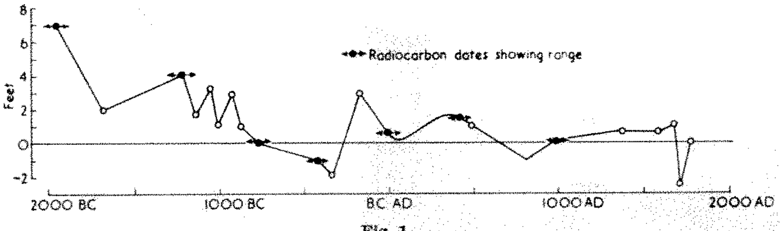

Fig. 1

Details will be published shortly in the *New Zealand Journal of Geology and Geophysics*.

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[1] Bakker, J. P., *Geologie en Mijnbouw.*, **16**, 232 (1954).

[2] Bennema, J., *Geologie en Mijnbouw.*, **16**, 254 (1954).

[3] Tavernier, R., and Moormann, F., *Geologie en Mijnbouw.*, **16**, 201 (1954).

Recognition of Folds and Faults on Rock Surfaces beneath the Sea

INFORMATION about the folding and faulting of the rock layers beneath the sea can be obtained by geophysical techniques, used in conjunction with sampling; but it is not easy to give a precise position of the geological boundaries on the floor, and only large faults can be inferred. The required detail can be obtained, however, by means of a geologists' asdic, where there are rock floors which consist of alternations of hard and soft beds and the former, being resistant to erosion, stand proud as ridges. In the English Channel, the Purbeck, Portland, Kimmeridge, Corallian beds and the ancient rocks occurring as shoals to east and west of Eddystone give good results, while Tertiary, Chalk and New Red Sandstone beds are too homogeneous to be suitable.

The method makes use of echo-sounding equipment, but instead of pointing the beam vertically beneath a ship it extends laterally and is tilted down and stabilized at about five degrees below the horizontal, so that echoes can be obtained from any ridges within a wide belt of floor parallel to the ship's course. The value of the method has already been described[1] and the equipment has been used on R.R.S. *Discovery II* to make reconnaisance surveys of sea-floors around southern Britain, with recent publication of some studies of sedimentary structures[2,3]. During these surveys rock outcrops of the type shown in Fig. 1 were found some miles south of the Dorset coast. The ship moved along a straight course (top of figure) covering two miles between the time marks, and examined a belt of sea-floor 800 yards wide, giving an exaggeration of scale of about four times for the range abeam. On this continuous plan view can be seen the dark bands of rock outcrops turning round in a gentle fold between the bottom right and left corners of the figure. In the upper part of the record a secondary beam which reached the sea floor beneath the ship gave a profile, in which an outcrop (black) terminates as a low cliff sloping away from the ship, at more than five degrees below the horizontal. The cliff marks the edge of a bed while the dip slope must be in the opposite direction and approximately at right angles to the line of outcrop of the bed. The continuity of any bed is interrupted by a parallel series of small faults, marked on the edge of the record with '*F–F*'. The pattern becomes increasingly blurred and vanishes as outcrops are progressively masked by loose sand at the right side of Fig. 1.

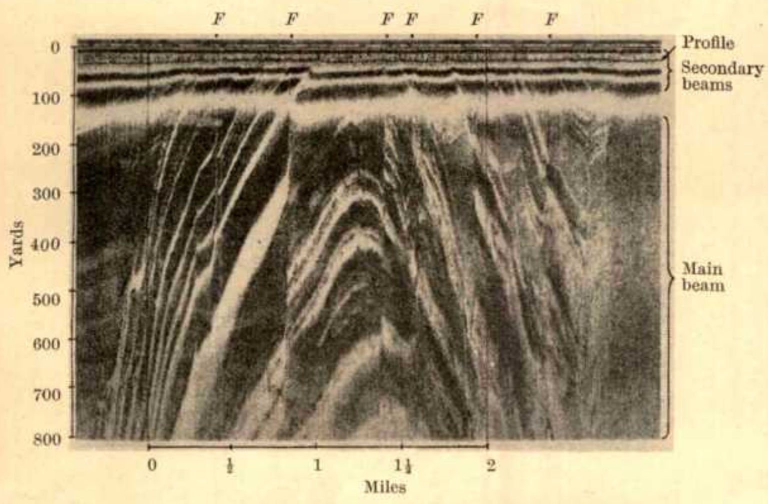

Fig. 1. A plan view of rock outcrops (black) on the sea floor showing part of a gentle fold and small fractures, '*F–F*'

A complete acoustic and sampling survey of this ground, off south-eastern Dorset, has since been made and an account, which is in preparation, will be published elsewhere with Dr. D. T. Donovan as co-author.

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[1] Chesterman, W. D., Clynick, P. R., and Stride, A. H., *Acustica*, **8**, 286 (1958).

[2] Stride, A. H., *J. Mar. Biol. Assoc. U.K.*, **38**, 313 (1959).

[3] Stride, A. H., *Dock and Harb. Authority*, **40**, 145 (1959).

Dissolution of Silicate Rocks

IN work on the determination of trace elements in silicate rocks[1], an attempt has been made to solve the inherent difficulties of achieving a rapid and efficient dissolution of large samples (up to 200 gm.) of silicate rocks and at the same time ensuring that the minimum quantities of reagents are used.

The classical treatment of rocks involves dissolving the sample in a sulphuric/hydrofluoric acid mixture or a perchloric/hydrofluoric acid medium, the sample being contained in a platinum dish. There are several disadvantages inherent in the use of platinum ware for these operations, in particular the extremely high price of platinum apparatus which is slowly destroyed by fuming with sulphuric acid. A further serious objection is the fact that no determinations of any of the platinoids can obviously be carried out in such a container.

Since I have been concerned mainly with the determination of very rare elements in silicate rocks, appreciably large samples have had to be used (on the average about 20 gm.). The dissolution of much smaller quantities of rock (say 1 gm.) presents few difficulties[2] but clearly a special technique had to be employed for these larger samples.

The dissolution technique presented here is the result of several years experience and depends on the use of 'Teflon' (polytetrafluorethylene) containers. This plastic possesses several very significant properties including an almost complete resistance to chemical attack by most reagents except molten alkali metals. It also possesses a marked freedom from warping or softening at temperatures up to 300° C. This material is expensive (a 150-ml. beaker costs about £6) but will certainly outlast platinum and is only a fraction of the cost of this metal.

It is hoped that a description of the method will be of assistance to other workers, particularly in the geochemical sphere, and will help them to reduce the tedium of the dissolution stage of chemical analyses on silicate rocks.

The technique described step-wise below is based on a sample weight of 40 gm. and can be carried out in a 150-ml. 'Teflon' beaker. If greater or lesser quantities are required, *pro rata* volumes of reagents must be used. (*a*) Weigh 40 gm. of finely powdered sample (100 mesh or greater) into a 150-ml. 'Teflon' beaker. (*b*) Add 10 ml. of aqua regia and stir into a paste with a 'Teflon' rod. (*c*) Very cautiously add 50 ml. of 'AnalaR' hydrofluoric acid in small amounts of about 3 ml. each. This is a hazardous operation and care must be taken to avoid frothing. (*d*) Add a further 40 ml. of aqua regia and take to dryness in a water-bath. Because of the low heat transmittancy of 'Teflon', the beaker should be suspended actually inside the water-bath with the top of the beaker protruding about ¼ in. above the top of the bath. (*e*) When the sample is dry (usually about 24 hr.), add 50 ml. of hydrochloric acid and transfer contents to a 250-ml. borosilicate glass beaker. Add 150 ml. of water and boil for about 5 min., allow to settle for ½ hr. and pour off the supernatant solution. (*f*) Transfer the residue back to the 'Teflon' beaker and again add 50 ml. of hydrofluoric acid and 50 ml. of aqua regia. Caution in adding reagents is not necessary in this step. (*g*) Again take to dryness in the water-bath and combine the residue with the original decanted liquid. (*h*) Add 50 ml. of sulphuric acid to the solution and fume to dryness to remove excess fluoride ions. This step is carried out in a borosilicate glass beaker and slight etching of the glass may occur due to liberation of fluoride ions. This degree of etching is slight and may be ignored unless a determination of alkali metals is being carried out, in which case careful blanks are necessary. (*i*) Add about 200 ml. of 2 *N* hydrochloric acid to the residue and 1 ml. of bromine. Boil for 5 min. and add sufficient hydrochloric acid (2 *N*) to adjust the total volume to about 1,250 ml.

In most cases the above treatment will result in a clear solution. Some rock types, however, will not dissolve completely with this treatment and an appreciable sediment may result. In such cases the sediment should be separated from the supernatant liquid and fumed with 10–15 ml. of sulphuric acid; the residue should then be added to the decanted liquid and, after boiling, dissolution should be complete.

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[1] Brooks, R. R., Ahrens, L. H., and Taylor, S. R., *Geochim. et Cosmochim. Acta*, **18**, 3 (1960).

[2] Edge, R. A., and Ahrens, L. H. (in the press).

METALLURGY

Corrosion Inhibition and the Zero Charge Potential of Metals

THE electrode potential of a mild steel specimen immersed in sodium chloride solutions has been measured in conditions of very rapid aeration produced by bubbling air vigorously over the specimen. Corrosion is rapid, but the potential does not fall to the values observed in conditions of moderate aeration. It rises to a maximum value that can be maintained for an appreciable time even though corrosion continues at a rapid rate. (This maximum potential must be related to the maximum rate of cathodic depolarization as limited by the rate of diffusion of oxygen across the boundary layer on the surface of the metal.) The relationships between potential, chloride concentration and surface preparation of the steel are specific and reproducible and will be described elsewhere. At lower rates of aeration, lower (less-positive) steady potentials can similarly be maintained. This method of potential stabilization was used to test the theory put forward by De[1] for inhibition in neutral and acid solution. De suggested that inhibitive anions are adsorbed preferentially on metal/solution interfaces that are positive to the zero charge potential (potential of electrocapillary maximum) of the metal, that is, he suggested, on cathodic surfaces in neutral solution. Evidence for such preferential adsorption has been found by Hoar and Holliday[2], by Antropov *et al.*[3] and others for inhibition in acid solutions; but so far as I am aware no direct evidence exists for its validity in neutral solutions.

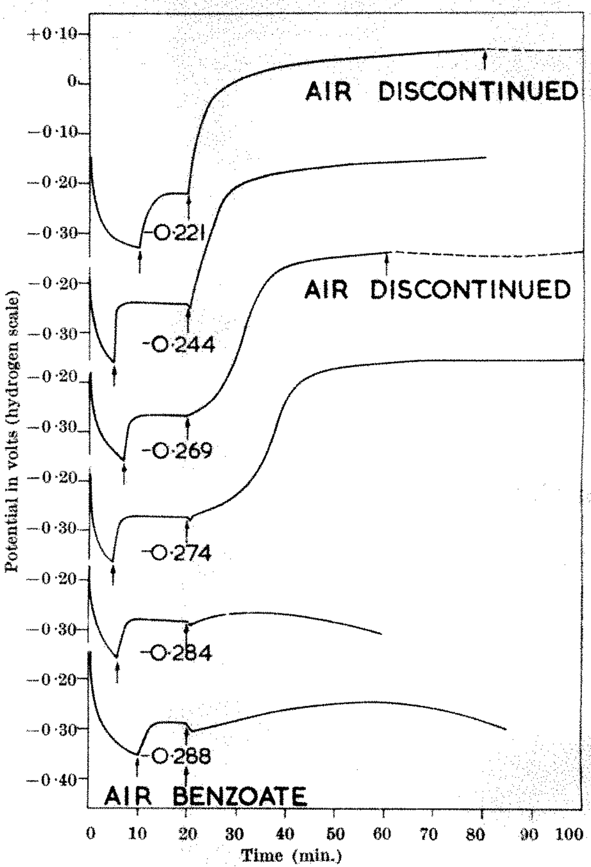

Fig. 1. Potential/time curves. Grit-blasted mild steel in 0·01 *M* sodium chloride

Grit-blasted specimens were suspended in 0·01 *M* sodium chloride (at approximately 20° C.), and after the potential had fallen to approximately −0·36 V. (hydrogen scale), which happens quickly in these conditions, a slow stream of air was passed through the solution (not over the specimen) at such a rate as to cause the specimen to attain a steady pre-determined potential. After 20-min. total immersion of the specimen, sodium benzoate was added, in sufficient amount to make the resulting concentration 0·1 *M*. From Fig. 1 it is seen that at potentials above −0·28 V. benzoate will induce inhibition; the higher the potential is above this value, the more readily does inhibition occur. Below this potential it will not induce inhibition. The conditions in all the solutions are precisely similar, the only variant being the potential reached by the specimen at the moment of addition of the benzoate. As the conductivity of the solution is high, the potentials of anodic and cathodic surfaces must be close together; hence adsorption presumably occurs over the whole surface irrespective of the anodic or cathodic nature of local areas.

If this hypothesis is correct, then the zero charge potential of steel in these solutions (0·01 *M* sodium chloride at *p*H 6) is approximately −0·28 V. (hydrogen scale). (Ayazyan[4] reports a value of −0·37 V. for iron in 0·001 *N* sulphuric acid.) Some corroboration that this value does correspond to the zero charge potential is provided by the growth of interference colour films on cathodically polarized surfaces; under similar conditions of *p*H, deposition of the positively charged colloidal corrosion product takes place at potentials just below this[5].

These results can now be used to interpret inhibitor mechanism in solutions that are in equilibrium with air, but not vigorously aerated. The primary function of oxygen (in all solutions of non-oxidizing inhibitors) is clearly to maintain the potential of the specimen above the zero charge potential for a sufficient time to enable the inhibitive anion to become adsorbed. De[6] showed that this took a measurable time (of the order of seconds) in the case of chromate ions. Upon immersion of the specimen, the potential will have a tendency to fall rapidly, the more so the higher the chloride concentration of the solution and the more reactive the metal surface (for example, grit-blasted in comparison with emeried); if the anion can become adsorbed quickly enough the fall in potential will be arrested and inhibition will ensue. For example, it was found that grit-blasted steel, in a stagnant solution of the same final composition as that of the solutions already used (0·01 *M* sodium chloride + 0·1 *M* sodium benzoate), remained protected, but only just; the potential fell slightly during the first 5 min. of immersion, then recovered. The 'fate' of the specimen will thus be decided by the relative speeds of two changes: the fall in potential (resulting from oxygen consumption and a decrease in anodic polarization) and the adsorption of anions. Aeration is not necessary in solutions of nitrite or chromate (or other oxidizing inhibitors), because these substances can raise the potential of the steel sufficiently to allow their own anions to become adsorbed.

This hypothesis explains several facts (hitherto unexplained) relating to inhibition by sodium benzoate. (*a*) Benzoate alone will not protect a badly rusted surface in stagnant conditions; in practice, the situation might be saved by a vigorous aeration 'boost'. (*b*) If benzoate is present in insufficient concentration to give inhibition, then general, but not localized, corrosion occurs. This may be ascribed to the utilization of the nearby oxygen in the early stages of breakdown, followed by a fall in potential, with consequent desorption of the rest of the benzoate anions. In highly aerated conditions, such as those produced by rapid movement, benzoate might no longer be a 'safe' inhibitor. With chromate or nitrite, even in stagnant solutions, the potential (except in case of excess chloride) tends to remain above the zero point owing to the oxidizing properties of these solutions. (*c*) The behaviour of steel in benzoate solution, when coupled to zinc or noble metals, was studied by Gilbert[7], who showed that (1) if steel and zinc were immersed separately in benzoate for 43 hr. and then connected, the potential of the couple was −0·21 V. (hydrogen scale), and the steel remained bright; (2) if steel and zinc in benzoate were connected from the outset, the steady potential of the couple remained in the region of −0·45 to −0·37 V.; that of galvanized steel remained at −0·35 V.; rusting of the steel took place in both cases; (3) no rusting occurred when steel was connected in benzoate solution to platinum, copper, or platinum with an externally applied current of 35 μamp., with the steel (1·5 cm. × 1·4 cm.) as anode; heavy rusting occurred if the current was reversed and the steel made the cathode. The potentials were not given but the results agree qualitatively with the hypothesis.

The potential of an immersed metal, relative to its zero charge potential, is clearly of fundamental importance in determining whether inhibition, due to adsorption of ions, is possible; in particular, a potential positive to the zero point is essential for inhibition by anions. Factors that determine this potential include the surface condition of the metal, the concentration of aggressive ions in the environment and the presence of oxygen or other cathodic depolarizers, contact with other metals or the passage of current.

Thanks are due to Dr. F. Wormwell for his interest and advice. This communication is published by permission of the Director of the National Chemical Laboratory.

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Feb. 8.

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[1] De, C. P., *Nature*, **180**, 803 (1957).

[2] Hoar, T. P., and Holliday, R. D., *J. App. Chem.*, **3**, 502 (1953).

[3] Antropov, L. I., Grigor'ev, V. P., and Petrenko, A. T., *Zh. prikl. Khim.*, **31**, 1497 (1958).

[4] Ayazyan, E. O., *Doklady Akad. Nauk. S.S.S.R.*, **100**, 473 (*Chem. Abst.*, **49**, 11533*a* (1955)).

[5] "Chemistry Research 1958", 30 (H.M.S.O., London, 1959) and unpublished results.

[6] Brasher, D. M., and De, C. P., *Nature*, **180**, 28 (1957).

[7] Gilbert, P. T., and Hadden, S. E., *J. App. Chem.*, **3**, 545 (1953).

Variation of Magnetic Moment with Composition in the Iron–Vanadium Sigma Phase

SINCE the structure of the sigma phase was established by Bergman and Shoemaker[1], it has been found in many binary alloys of transition elements (see, for example, Haworth and Hume-Rothery[2]). Where the phase boundaries have been established, the width of the range of composition varies from less than 1 per cent in the molybdenum–cobalt system to about 20 per cent in the iron–vanadium system. On the basis of the positions of the phase in various binary alloy phase diagrams, it has been suggested that the sigma phase is some kind of electron compound (see, for example, Kasper[3]). Greenfield and Beck[4] concluded that while the sigma phase might be associated with a particular average number of vacancies in electronic '*d*' states per atom, no self-consistent scheme could be developed to reconcile both theory and experiment. Nevertheless, the possibility that '*d*' state vacancies might play a part in stabilizing the structure suggests that magnetic evidence would prove of value. Consequently, in a survey of many binary and ternary sigma phase alloys, Nevitt and Beck[5] concluded that all were ferromagnetic, having Curie points in the temperature range 50–200° K. No measurements had been made,

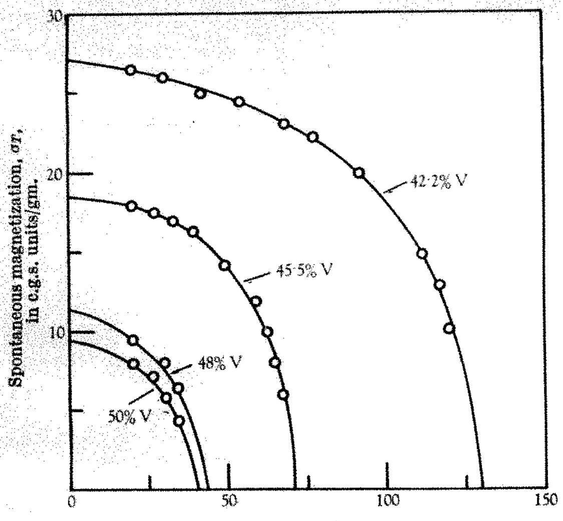

Fig. 1. Variation of spontaneous magnetization, σ_T, with temperature for the four iron–vanadium sigma phase alloys

however, of the mean magnetic moment per atom (p_B) of any ferromagnetic sigma phase, or of its variation, if any, with composition, and so the present work was undertaken, the iron–vanadium system being chosen because of the comparatively wide range of composition over which the sigma phase extends.

The alloys were prepared by melting together appropriate amounts of iron and vanadium in an electric arc furnace under an atmosphere of argon at a pressure of about 40 cm. of mercury. The iron used was B.I.S.R.A. iron of 99·96 per cent purity, and the vanadium was supplied by New Metals and Chemicals, Ltd., and was of 99·5 per cent purity. The sigma phase was precipitated by heating for four days *in vacuo* at 1,050° C., the presence of the phase then being confirmed by X-ray diffraction. Measurements of the magnetization were then made at temperatures between 20° K. and the Curie point and in applied magnetic fields of between 4 and 16 kilo-oersteds, using a Sucksmith ring balance[6]. The linear part of the magnetization curves at the constant temperature, T, were extrapolated to zero field to determine the spontaneous magnetization at that temperature, σ_T. The spontaneous magnetization at absolute zero, σ_0, and the Curie point, θ_f, were deduced using methods which have become standard in this laboratory (see Crangle[7]). The results are shown in Fig. 1 and Table 1, and from them it can be seen that there is a decrease in magnetic moment and Curie point with increasing vanadium content. A linear extrapolation of the curve of magnetic moment against composition goes to zero moment at a composition corresponding to about 53 per cent vanadium although the composition limit of the sigma phase is about 57 per cent vanadium. The Curie points are considerably lower than that of the α-phase of the same composition as deduced from Fallot[8], so ruling out the possibility that the ferromagnetism is due to retained alpha phase.

Table 1

Composition percentage V (by atoms)	σ_0 (c.g.s. units/gm.)	Curie point (° K.)	p_B (Bohr magnetons)
42·2 ± 0·5	27·1	130	0·261
45·5 ± 0·1	18·4	72	0·177
48·0	11·5	44	0·110
50 ± 2	9·5	40	0·091

There would seem to be two possible interpretations of the results.

If the magnetic moment of the sigma phase is due to a net unbalance of positive and negative spins in a common '*d*' band, then there is a redistribution of spins in the band as the composition changes, leading to a balance of spins at 53 per cent vanadium. The phase does not, however, cease to be stable at this point.

The alternative interpretation is that there is a ferri-magnetic arrangement of magnetic ions in the structure which alters with composition. This would seem to be the more probable interpretation, particularly in view of the fact that the curves of inverse susceptibility against temperature above the Curie point are slightly concave to the temperature axis.

The compositions quoted in Table 1 are based on the amount of each constituent present before melting, and the errors estimated from the loss in weight during melting.

Further work is in progress on the iron–vanadium system, and also on other sigma phase alloys.

It is a pleasure to record my thanks to Prof. W. Sucksmith for his advice and for his continued interest in the work.

The alloy quoted as containing 48 per cent vanadium was kindly supplied by Dr. B. R. Coles.

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[1] Bergman, G., and Shoemaker, D. P., *Acta Cryst.*, **7**, 857 (1955).
[2] Haworth, C. W., and Hume-Rothery, W., *Phil. Mag.*, **3**, No. 33, 1013 (1958).
[3] Kasper, J. S., "Theory of Alloy Phases" (A.S.M.), 264.
[4] Greenfield, P., and Beck, P. A., *J. Metals*, **8**, 265 (1956).
[5] Nevitt, M. V., and Beck, P. A., *Trans. Amer. Inst. Mining and Metallurgical Eng.*, **203**, 669 (1955).
[6] Sucksmith, W., *Proc. Roy. Soc.*, A, **170**, 551 (1939).
[7] Crangle, J., *Phil. Mag.*, **46**, 499 (1955).
[8] Fallot, M., thesis, Strasbourg (1935).

CHEMISTRY

Conductivity of Aqueous Sodium Stannate Solution

IN view of the attention which has recently been paid to the chemistry of silicates and germanates[1,2] the following results obtained during an investigation of the properties of sodium stannate solutions are of interest.

When sodium stannate, prepared by Zocher's method[3], is dissolved in water at 25·0° C. there is an immediate increase in *p*H, after which the solution becomes slowly more alkaline, reaching a steady limiting value after about one week. In order to follow this reaction the variation in conductivity with time was measured using an electrodeless capacity-coupled cell[4]. Over the whole of the concentration-range studied the specific conductance increased regularly with time and reached a limiting value after seven days. The specific and equivalent conductances at zero time were obtained by graphical extrapolation of the conductivity-time plots; the results are given in Table 1.

Table 1. Equivalent Conductance of Sodium Stannate Solutions at Zero Time

Molar concentration sodium stannate	0	$1\cdot035 \times 10^{-4}$	$1\cdot179 \times 10^{-4}$	$1\cdot735 \times 10^{-4}$	$2\cdot639 \times 10^{-4}$
Equivalent conductance (ohm^{-1} cm.^{2})	111·8	110·7	109·0	107·0	106·4
Molar concentration sodium stannate	$5\cdot485 \times 10^{-4}$	$7\cdot018 \times 10^{-4}$	$1\cdot051 \times 10^{-3}$	$1\cdot527 \times 10^{-3}$	$3\cdot553 \times 10^{-3}$
Equivalent conductance (ohm^{-1} cm.^{2})	105·0	103·1	100·3	99·5	93·9
Molar concentration sodium stannate	$4\cdot896 \times 10^{-3}$	$5\cdot216 \times 10^{-3}$	$6\cdot993 \times 10^{-3}$	$7\cdot564 \times 10^{-3}$	$1\cdot246 \times 10^{-2}$
Equivalent conductance (ohm^{-1} cm.^{2})	92·7	91·4	91·1	88·6	85·6

The equivalent conductivity at infinite dilution, 111·8, was obtained from a Kohlrausch plot; on correction for the effect of hydrolysis has been made. The difference between this value and the value of 156 obtained by Bellucci and Parravano[5] for potassium stannate is greater than the theoretical one. Bellucci and Parravano, however, only noted an increase in conductivity with time in their most concentrated solution, so it is possible that their potassium stannate was impure.

The increase in conductivity with time and the final attainment of a limiting value shows that the species initially present are reacting at a measurable rate. By means of *p*H measurements it has been shown that for approximately the first 70 per cent of the reaction the increase in specific conductivity is a direct measure of the increase in hydroxyl ion concentration of the solution. The rate of production of hydroxyl ions is a first or pseudo-first order reaction with a rate constant of $5\cdot3 \times 10^{-2}$ hr.$^{-1}$. The hydrolysis of sodium stannate is thus seen to occur in at least two stages: first, a fast reaction, typified by the immediate alkaline reaction of a freshly prepared stannate solution, and, secondly, by a slow first or pseudo-first order formation of more hydroxyl ions. Unfortunately, there are insufficient data available at present to make a more detailed mechanistic analysis of these reactions; but in view of the fact that both silicates and germanates form polynuclear hydrolysis products it seems likely that a similar explanation would apply in the case of stannate solutions.

I wish to thank the Superintendent of the Admiralty Materials Laboratory for permission to publish this work.

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[1] Igri, N., Abstracts International Conference on Co-ordination Chemistry, London, 1959.

[2] Everest, D. A., and Harrison, J. C., *J. Chem. Soc.*, 2178 (1959).

[3] Zocher, H., *Z. anorg. Chem.*, **112**, 1 (1920).

[4] Bromley, D. E., and Good, P. T., *J. Sci. Instr.*, **36**, 326 (1959).

[5] Bellucci, J., and Parravano, N., *Atti. Accad. Lincei*, **13**, 307 (1904).

Toxicity of Plastics

In view of the ever-increasing use of plastics in contact with foodstuffs and like materials and the enormous amount of work done to establish their suitability for such uses, second thoughts on materials condemned without actual test might be advisable. In this respect I would like to direct attention to the results of a series of tests carried out in the laboratories of Bakelite, Limited.

We were asked to investigate the leaching-out of lead compounds from lead-stabilized polyvinyl chloride compositions in contact with a constant volume of hard and soft water. These tests were carried out by storing 6 litres of water at room temperature in tanks 18 in. × 12 in. × 5 in. deep made from the sample material by welding press-polished sheet and, after a period of 18 months contact with the water, determining the total lead content of the waters. It was shown that, with a compound stabilized with 3·77 per cent basic lead phosphite and lubricated with 0·94 per cent basic lead stearate, 0·5 part per million lead were extracted by soft water and no lead by hard water; while a compound, containing only 3·81 per cent dibasic lead phosphite, had none of its lead extracted by either hard or soft water. Thus it appears that only some types of lead compound are extracted from these compositions by water.

Although the test specimens employed and the test methods used are not those specified by the British Plastics Federation in its Toxicity Report No. 40, dated 1958, nevertheless, it is felt that the results are of considerable interest to the industry as a whole. If the total amount of lead extracted from a sample over a period of 18 months is only 0·5 part per million, then surely such a material would be satisfactory for use in pipes in contact with flowing water. In the event of water being stagnant in the pipes up to 18 months it could only produce a lead concentration of 0·5 part per million. On the other hand, formulations can be made which contain lead compounds, for example, lead phosphite, from which no lead is extracted either by hard or soft water. These results could, therefore, be considered good evidence in favour of allowing a formulation containing lead compounds to be employed in contact with water in conformity with Continental practice.

In conclusion, I suggest that polyvinyl chloride compositions should not necessarily be condemned as being toxic just because they contain toxic substances, but should be considered in the light of the applications for which they are intended to be used.

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Separation Factors in Gas Chromatography

It is generally accepted among those using gas chromatography that for most separations the use of lower temperatures increases the separation factor, though, usually owing to the time taken at low temperatures, there is a limit to the use to which this effect can be put. Capillary columns can, however, be made extremely fast in their operation, and hence low temperatures can be used for separations which normally require temperatures at least 70° C. higher.

The classically difficult separation in gas chromatography is that of *meta*- and *para*-xylene. Although nowadays this separation is relatively commonplace owing to the use of capillary columns and selective stationary phases, an interesting separation was made in the course of routine testing at our laboratory which illustrates the effect of low-temperature operation. In this case, the stationary phase was a non-

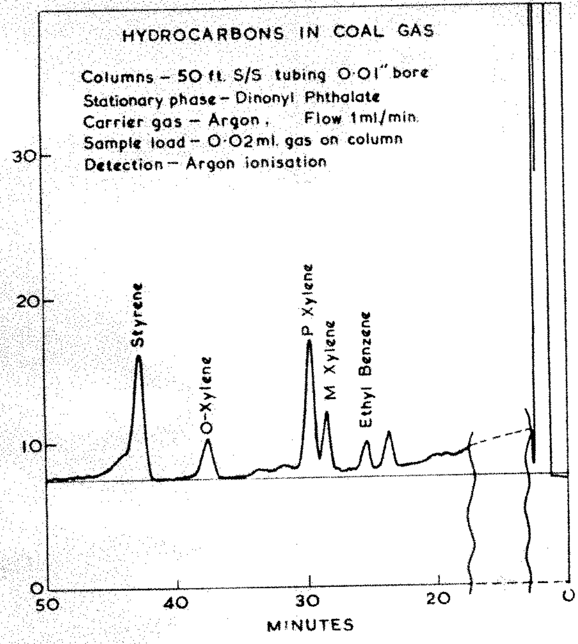

Fig. 1

selective one, dinonyl phthalate. At room temperature, these two isomers are easily separated, the separation factor being 1·05. A typical chromatogram is shown in Fig. 1. No separation is obtained at 80° C., although the plate number remains the same. It is clear that the principal separating factor with these two substances is their difference in vapour pressure, and that the difference is much greater at room temperature than it is at more elevated ones.

It is probable that the large increase in separation factor towards these really low temperatures is a general one.

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BIOCHEMISTRY

Hepatotoxic Activity of Semi-Synthetic Analogues of Pyrrolizidine Alkaloids

MANY of the natural pyrrolizidine alkaloids isolated from various plants are not only acutely hepatotoxic, but are also able to produce subacute and chronic liver lesions in rats when given in an appropriate single dose. The most striking feature of the lesions are the cytological abnormalities of the liver parenchymal cells, suggesting that the alkaloids may be interphase mitotic poisons[1]. The hepatotoxic activity of these alkaloids, especially in relation to their basic moieties, has been discussed[2]. Though their structures vary greatly, yet one feature is common to all, namely, the branching of the carbon-chain of their acidic moieties, the necic acids.

Such structures could in the course of the metabolic degradation in the animal body yield isoprenoid units, or their homologues. In view of the finding that isoprenoid units, derived from mevalonic acid (3 : 5-dihydroxy-3-methyl-valeric acid), are incorporated *in toto* into squalene, the precursor of cholesterol[3], and probably into steroidal hormones, and other polyprenoids, it is conceivable that the hepatotoxic pyrrolizidine alkaloids interfere in some way with the biosynthesis of some such vital cell constituents. The fact that young, immature animals are more susceptible than adults, and males more than females, to the action of pyrrolizidine alkaloids[4] suggests also that steroidal hormones may be implicated.

Table 1. HEPATOTOXIC ACTIVITY IN RATS

Compound	Dose range (mgm./kgm. body-weight)	Cytological abnormalities of liver
Retronecine hydrochloric acid	600–1,000	–
Monocrotalic acid lactone (sodium salt)	400–1,300	–
Tiglic acid (sodium salt)	60–1,670	–
Tiglic amide	140– 600	–
Senecioic acid (sodium salt)	400–2,500	–
Senecioic amide	170– 500	–
Di-*n*-butyryl retronecine	175–1,300	–
Di-*n*-valeryl-retronecine	480– 730	–
Di-*iso*-valeryl-retronecine	300– 830	+ +
Di-tigloyl-retronecine	100– 500	+ +
Di-senecioyl-retronecine	200– 525	+ + +

Yet, the free necic acids obtained by hydrolysis of the natural alkaloids are not hepatotoxic, nor are synthetic acids with an isoprenoid structure such as senecioic or tiglic acids, or their amides (Table 1).

It was thought desirable to try to prepare semi-synthetic analogues of pyrrolizidine alkaloids with a series of related straight and branched-chain, monocarboxylic acids, in order to evaluate the significance of the branching of the carbon chain of the acidic moieties in relation to their hepatotoxic activity.

The amino-alcohol used for this purpose was natural retronecine, obtained by hydrolysis of monocrotaline (the main alkaloid of *Crotalaria retusa* L.), according to the procedure of Adams and Rogers[5]. This was esterified by the action of the respective acid chlorides, yielding mainly diesters. These were oily liquids, and were purified via their crystalline picrates. The preparation of di-senecioyl-retronecine may serve as an example. Retronecine hydrochloride was heated with an excess of redistilled senecioyl chloride, on a water-bath for 15 min.; the excess of the chloride was decomposed with alkali and the ester, extracted with ether, was isolated as a crystalline picrate, yellow needles, m.p. 115° C., from ethanol (found : C, 52·9 ; H, 5·24 ; N, 10·3 per cent : $C_{24}H_{28}N_4O_{11}$ requires C, 52·6 ; H, 5·1 ; N, 10·2 per cent). The diester, a colourless viscous oil, was recovered from the picrate by shaking with aqueous ammonia and extracting with ether. It formed a crystalline hydrochloride, colourless needles from acetone-ether, m.p. 126° C. (found : C, 60·5 ; H, 7·37 ; N, 4·3 per cent : $C_{18}H_{26}NO_4Cl$ requires C, 60·7 ; H, 7·3 ; N, 3·9 per cent).

For biological testing the respective compounds were dissolved, by the addition of equimolecular amounts of dilute hydrochloric acid, or in water when present as hydrochlorides, and various, single doses were given by stomach tube or by intraperitoneal injections to weanling Wistar rats. The animals which did not die within a few days were allowed to survive for at least 1–2 months, before they were killed and examined. The livers, lungs and sometimes other organs were then fixed and processed in the usual way for microscopic examination. The presence of significant numbers of characteristic large parenchymal liver cells was taken as evidence of hepatotoxic activity.

The diesters, when tested by intraperitoneal injections, caused convulsions and death of the rats, with

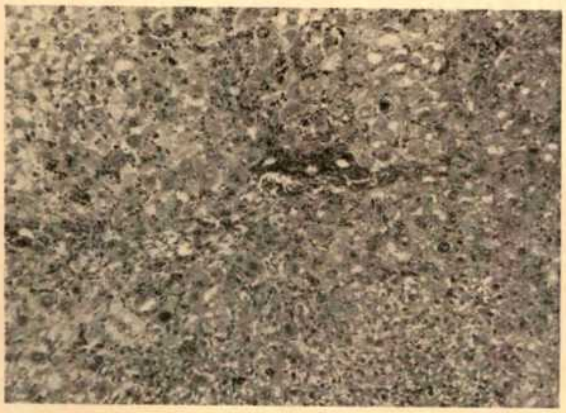

Fig. 1. Liver of a male rat, killed 3 months after a single dose of di-senecioyl-retronecine, 450 mgm./kgm. body-weight, showing great variation in the size of parenchymal cells and infiltration of portal areas (hæmatoxylin and eosin, × 100)

accompanying lung œdema, after doses much smaller than (about 1/5) those which produced similar effects by the oral route. Indeed, only some of the compounds were given in high enough oral doses to induce convulsions, which were then less intense and transitory, so that the animals only died a few days after the treatment.

Subacute liver lesions and cytological abnormalities indistinguishable from those seen after the natural pyrrolizidine alkaloids were produced by the diesters of retronecine with tiglic and senecioic acids, administered orally (Fig. 1). The doses required to produce these lesions were of the order 250–500 mgm./kgm. body-weight (Table 1). These doses are higher than those of the natural alkaloids, whether the cyclic or the open esters[2], which produce similar lesions. This may be due to the use of the synthetic products as such, without trying to resolve them into optically active isomers.

The liver lesions produced by di-*iso*-valeryl-retronecine were comparable, but no evidence of abnormalities were seen in the livers of rats given di-*n*-butyryl-, and di-*n*-valeryl-retronecine, even after doses as high as 1,300 mgm. and 730 mgm./kgm. body-weight, respectively.

These results are consistent with the view that branching of the carbon-chain of the acidic moieties is necessary for hepatotoxic activity of this class of compounds. With the possibility now open for the synthesis of appropriately labelled hepatotoxic analogues of pyrrolizidine alkaloids, further work on the mechanism of their toxic action can be pursued.

The experimental details of this work will be published elsewhere. We wish to thank Dr. C. D. Adams, Department of Botany, Achimota, and Dr. R. A. Galley, Tropical Products Institute, London, for their kind help in obtaining large quantities of *Crotalaria retusa* from Ghana, as a source of monocrotaline, and Dr. J. M. Barnes for his stimulating interest.

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[1] Schoental, R., and Magee, P. N., *J. Path. Bact.*, **78**, 471 (1959).

[2] Schoental, R., *Nature*, **179**, 361 (1957).

[3] Cornforth, J. W., Cornforth, R. H., Popjak, G., and Gore Youhotsky, I., *Biochem. J.*, **69**, 146 (1958).

[4] Schoental, R., *J. Pat[illegible]. Bact.*, **77**, 485 (1959).

[5] Adams, R., and Rogers, E. F., *J. Amer. Chem. Soc.*, **61**, 2815 (1939).

Formation of L-Xylosone from Ascorbic Acid

DURING a study of the carbonyl compounds produced during cider fermentations an immediate precipitate (*A*) was obtained at room temperature on addition of dinitrophenylhydrazine to a sulphited cider (that is, a cider obtained from a juice to which 150 parts per million sulphur dioxide had been added after milling and pressing the fruit). A similar precipitate was formed in a sulphited Bramley Seedling juice which had failed to ferment, but no appreciable precipitate was noted in non-sulphited fermentations. The derivative (*A*) on a paper chromatogram gave a blue colour with sodium hydroxide solution which indicated the possibility of osazone formation. Owing to the difficulty of purifying the 2 : 4-dinitrophenylhydrazine derivative, the phenylhydrazine derivative (*B*) was prepared from a sulphited cider at room temperature.

Paper chromatography, that is, examination of derivative *B* by running on the same untreated No. 1 Whatman paper and on borate-treated paper with the phenylosazones of glucose, xylose and arabinose in dioxane–heptane–water (2 : 1 : 1), suggested that *B* was xylosazone.

Table 1

	R (arabinosazone) values	
	Untreated paper	Borate-treated paper
L-Arabinosazone	1·00	1·00
D-Xylosazone	1·00	0·25
Isolated derivative (*B*)	1·00	0·25
D-Glucosazone	0·37	0·00

Derivative *B* was purified by elution from an alumina column[1] with alcohol–water (9 : 1) after washing with benzene, benzene–ether (1 : 1) and ether. After evaporating to dryness *B* was recrystallized from methanol–water and had a melting point 164–165°, identical with that of D-xylosazone. A mixed m.p. of *B* and D-xylosazone gave m.p. 196°, which indicated that compound *B* was L-xylosazone. The m.p. of L-xylosazone made from a commercial sample of L-xylose was 168–169° and a mixed m.p. of this derivative with compound *B* gave 166–167°. Compound *B* was, therefore, identified as L-xylosazone and the substance present in the cider as L-xylosone.

The origin of this substance is of some interest. Recent work has shown that sugars may be oxidized to osones by the action of bisulphite[2]. However, the xylose present in the cider-apple juices is D-xylose (shown by m.p. and mixed m.p. of isolated xylosazone and of an authentic specimen). A more likely source of L-xylosone is L-ascorbic acid, which has been shown to occur in appreciable amounts in cider apple juices[3]. During the milling and pressing processes prior to the addition of sulphur dioxide it would be expected that much L-ascorbic acid would be converted to L-dehydroascorbic acid, also dehydroascorbic acid may be present in amounts similar to ascorbic acid in apples[4]. Opening up of the lactone ring followed by decarboxylation would yield L-xylosone.

A sample of ascorbic acid was oxidized to dehydroascorbic acid with iodine solution and just decolorized with sulphur dioxide solution. Sufficient sulphur dioxide solution was then added to combine with the carbonyl groups. After standing three days at room temperature the solution on addition of 2 : 4-dinitrophenylhydrazine rapidly gave a precipitate in the cold. A larger precipitate was obtained after boiling the solution for 30 min. Addition of phenylhydrazine and acetic acid gave an osazone in the cold with m.p. 164–165° unchanged on mixing with L-xylosazone isolated from cider. Standing or heating L-dehydroascorbic acid with sulphur dioxide therefore formed L-xylosone.

Previous work has shown that ascorbic acid is oxidized by perinaphthindan-2 : 3 : 4 trione hydrate at 45° to dehydroascorbic acid, which then undergoes hydrolysis and decarboxylation to yield L-xylosone[5]. Similarly, oxidation with ninhydrin[6] and alloxan[7] was followed by decarboxylation with the probable formation of L-xylosone. The formation of L-xylosone from dehydroascorbic acid in the presence of sulphur dioxide is of interest in view of the high proportion of dehydroascorbic to ascorbic acid in apples, pears and cherries[4] and of the use of sulphur dioxide as a preservative in other food products.

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[1] Barry, V. C., and Mitchell, P. W. D., *J. Chem. Soc.*, 4020 (1954).

[2] Ingles, D. L., *Chem. and Indust.*, 1045 (1959).

[3] Kieser, M. E., and Pollard, A., *Nature*, **159**, 65 (1947).

[4] Koch, J., and Bretthauer, G., *Landwirtsch. Forsch.*, **9**, 51 (1956).

[5] Moubasher, R., *J. Biol. Chem.*, **176**, 529 (1948).

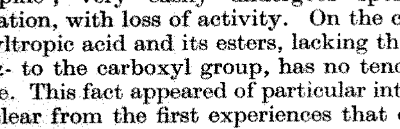
[6] West, E. S., and Rinehart, R. E., *J. Biol. Chem.*, **146**, 105 (1942).

[7] Nath, M. C., and Bhattathiry, E. P. M., *Nature*, **176**, 787 (1958).

Anticholinergic Activity of Tropine-α-Methyltropate

THE synthesis of the new α-methyltropic ester of tropine and the isolation of the two optically active isomers has been recently described by Melone, Vecchi, Pagani and Testa[1].

These compounds (DL, (+)- and (−)-tropine-α-methyltropate) were supposed to display an anticholinergic activity possibly different from atropine itself (especially as far as the duration of action is concerned). In fact, the above-quoted authors, as well as Foster and Ing (who attempted to synthesize the same compounds[2]), had suggested that the esters of α-methyltropic acid, being very difficult to hydrolyse, should lead to quite stable compounds. Moreover, the natural occurring (−)-atropine, practically responsible for most of the action of atropine[3], very easily undergoes spontaneous racemization, with loss of activity. On the contrary, α-methyltropic acid and its esters, lacking the hydrogen in α- to the carboxyl group, has no tendency to racemize. This fact appeared of particular interest, as it was clear from the first experiences that only the (−)-isomer of tropine α-methyltropate possesses a significant anticholinergic activity.



Tropine-α-methyltropate

The stability of tropine α-methyltropate was believed to exercise an influence not only on the degree but also upon the duration of the pharmacological effects, as it is well known that the natural alkaloid, atropine, is destroyed by mammalian tissues, mostly through enzymatic hydrolysis of the ester to tropine (or tropanol) and tropic acid[4].

This communication concerns the results of a preliminary pharmacological investigation, carried out on (−)- and (+)-tropine α-methyltropate hydrochlorides.

The anticholinergic activity of the two compounds was evaluated and compared with that of atropine sulphate, on isolated rat gut and on the cardiovascular response of the dog to intravenously injected acetylcholine.

Acute intravenous toxicity was studied in *CF*-1 mice.

In isolated rat intestine the products were added to the oxygenated Tyrode solution of the bath chamber in fixed volume of 0·2 ml., 1 min. before the introduction of acetylcholine chloride. The recorded effects were evaluated as reduction of the contractile response to acetylcholine.

In preliminary tests it was demonstrated that (−)- and (+)-isomers and atropine display an anticholinergic activity qualitatively similar.

The concentration/activity curves of the three drugs were found to be parallel, thus allowing the estimation of relative potency. Three comparisons were carried out: between atropine sulphate and the (−)-α-methyl derivative hydrochloride; between atropine sulphate and the (+)-α-methyl derivative hydrochloride; and finally between the two optical antipodes. By calculating the ratios between equally effective concentrations (that is, the concentrations effective in reducing to 50 per cent the response of acetylcholine afterwards administered) the following potency ratio resulted:

Atropine/(−)-methylatropine (No. of experiments, 8)	1–2
Atropine/(+)-methylatropine (No. of experiments, 7)	1–0·04
(−)-Methylatropine/(+)-methylatropine (No. of experiments, 9)	1–0·02

The average effective concentration of atropine sulphate was found to be within a range of 0·016–0·012 μgm./ml. The average effective concentration of (+)-α-methylatropine was found to be 0·36–0·22 μgm./ml., while for (−)-α-methylatropine it was found to be 0·0074–0·0038 μgm./ml.

In dogs anæsthetized with sodium pentobarbitone the ability of (−)-methylatropine and atropine sulphate to prevent the fall in blood pressure caused by intravenous administration of 2 μgm. of acetylcholine was compared and the duration of the inhibitory effect especially considered. Each of the two products was injected intravenously in doses of 5 μgm./kgm. and acetylcholine administered before and 2, 6, 16, 26, 36 and 46 min. after the dosage until the fall in blood pressure produced by acetylcholine was returned to pre-treatment level. Six dogs were employed for these experiments. Three of them received the two products alternately. The three other dogs received the two drugs on the basis of a random schedule. The dose-level of 5 μgm./kgm. was chosen after preliminary assays that demonstrated this to be approximately the average effective dose for the two drugs.

In Table 1 the results of these experiments are briefly summarized.

The *LD*50 of (−)-atropine-α-methyltropate after intravenous administration in mice was found to be 102 mgm./kgm. (confidence limits $P = 0{\cdot}05$: 89·57–116·3) (according to Litchfield and Wilcoxon[5]), while for atropine the intravenous *LD*50 is 76·7 mgm./kgm. (confidence limits: 71·8–81·9). Also acute toxicity of both drugs in mice after intravenous administration gave the same result.

Table 1

Compound	No. of experiments	Dose (μgm./kgm.)	Average inhibition (per cent) of the depressor effect of acetylcholine (2·0 μgm./kgm.) after					
			2 min.	6 min.	16 min.	26 min.	36 min.	46 min.
Atropine sulphate	18	5	81·00 ± 7·62	59·50 ± 6·13	41·16 ± 7·32	27·83 ± 8·48	13·20 ± 7·33	3·80 ± 1·64
(−)-Methylatropine	15	5	81·20 ± 5·58	51·60 ± 10·4	34·00 ± 7·72	23·25 ± 9·24	13·00 ± 8·19	10·75 ± 8·29

Our preliminary experiments indicate that between the two optical antipodes of the new derivative tropine-α-methyltropate, the (−)-isomer shows *in vitro* a cholinergic blocking activity about fifty times greater than the corresponding (+)-isomer. This difference is substantially of the same order as between the optical isomers of the naturally occurring alkaloid atropine. Bovet and Bovet-Nitti[3] report that *l*-hyosciamine is ten to one hundred times more active than the *d*-isomer, according to different evaluations.

In *in vitro* experiments (−)-atropine-α-methyltropate hydrochloride shows an anticholinergic activity twice as great as atropine sulphate. In the dog, however, atropine sulphate and (−)-tropine-α-methyltropate hydrochloride were found equivalent in their ability to prevent the depressor effect of acetylcholine on blood pressure. Although there is no experimental evidence to support the hypothesis that the great resistance of (−)-atropine-α-methyltropate to hydrolysing agents could implicate a greater duration of pharmacological activity, the preliminary results herein reported seem to indicate that (−)-tropine-α-methyltropate is a drug of potentially practical value.

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[1] Melone, G., Vecchi, A., Pagani, G., and Testa, E., *J. Org. Chem.* (in the press).

[2] Foster, R., and Ing, H. R., *J. Chem. Soc.*, 938 (1956).

[3] Bovet, D., and Bovet-Nitti, F., "Structure et Activité des Médicaments du Système Nerveux végétatif", 500 (S. Karger, Basle, 1948).

[4] Goodman, L. S., and Gilman, A., "The Pharmacological Basis of Therapeutics", second edit., 551 (The Macmillan Co., New York, 1956).

[5] Litchfield, J. T., and Wilcoxon, F., *J. Pharm. Exp. Therap.*, **96**, 99 (1949).

11α-Hydroxylation of Steroids by *Streptomyces* Species

Extensive studies[1] in the past few years have shown that the ability to transform the steroid molecule is widespread among various micro-organisms, and the types of reaction are numerous. Among them, 11α-hydroxylation is a quite common reaction with fungi[1,2]. More recently, it was found that two strains of bacteria have introduced the hydroxy group in progesterone[3] and Reichstein's[4] compound *S* at the 11α-position. However, with Actinomycetes this 11α-hydroxylation has not been reported, while 11β-hydroxylation of compound *S* by *Streptomyces radiae*[5] has been shown.

In the present communication we wish to report that microbiological hydroxylation of progesterone and Reichstein's compound *S* in the 11α-position and the 6β-position has been carried out by unidentified *Streptomyces* species (*Ac*-103) isolated from soil. In our experiment, *Streptomyces* sp. (*Ac*-103) was incubated in a medium containing 1 per cent glucose, 1 per cent peptone, 1 per cent meat extract and 0·3 per cent sodium chloride. After incubation at 28° for a period of 48 hr. on a reciprocating shaker, 1 gm. of the steroid substrate dissolved in methanol was added to 2 l. of medium and the incubation was continued for 48 hr. The culture filtrate was extracted with ethyl acetate ester and the extracts were concentrated to dryness *in vacuo* at 30°. The residue was redissolved in benzene or ethylene dichloride and the solution was chromatographed on an alumina or 'Florisil'.

When progesterone was used as a substrate in the incubation with this micro-organism, 6β,11α-dihydroxyprogesterone was isolated as the insoluble matter in benzene of the condensed residue of extract. Crystallization of this insoluble matter from acetone afforded 400 mgm. of 6β,11α-dihydroxyprogesterone (I), m.p. 234–240°, $[\alpha]_D + 110°$ (methanol), $\lambda_{\mathrm{max.}}^{\mathrm{MeOH}}$ 236 mμ ($\varepsilon = 14,200$), $\lambda_{\mathrm{max.}}^{\mathrm{KBr}}$ 3,420 (OH), 1,695 (20-CO), 1,660 and 1,615 (Δ^4-3-CO) cm.$^{-1}$. The diacetate of (I) prepared with acetic anhydride and pyridine melts at 150–153°, $[\alpha]_D + 70°$ (chloroform). The infra-red spectra of these compounds agreed with those of authentic samples. Then, the benzene solution filtrate was chromatographed on alumina and benzene–ether–chloroform gradient elutions were used for the separation of other oxygenated steroids. In the first fractions a small amount of progesterone was obtained, and in the second fractions 6β-hydroxyprogesterone was obtained. The crystallization of the second fractions from acetone gave 100 mgm. of 6β-hydroxyprogesterone (II), m.p. 172–173°, $[\alpha]_D + 98°$ (chloroform), $\lambda_{\mathrm{max.}}^{\mathrm{MeOH}}$ 236 mμ ($\varepsilon = 13,700$), $\lambda_{\mathrm{max.}}^{\mathrm{KBr}}$ 3,450 (OH), 1,710 (20-CO), 1,669 and 1,620 (Δ^4-3-CO) cm.$^{-1}$.

When Reichstein's compound *S* was used as a steroid substrate, the 6β-hydroxylated product and 11α-hydroxylated product were obtained by column chromatography on 'Florisil' using ethylene dichloride–acetone gradient elutions. The product obtained by recrystallization of the first fractions from acetone was 320 mgm. of 6β,17α,21-trihydroxy-4-pregnene-3,20-dione (III), m.p. 230–235°, $[\alpha]_D + 51°$ (methanol), $\lambda_{\mathrm{max.}}^{\mathrm{MeOH}}$, 238 mμ ($\varepsilon = 13,300$), $\lambda_{\mathrm{max.}}^{\mathrm{KBr}}$ 3,420 (OH), 1,710 (20-CO), 1,665 and 1,615 (Δ^4-3-CO) cm.$^{-1}$. The 6,21-diacetate melts at 190–195°, $[\alpha]_D +$ 60° (methanol). Crystallization of the second fractions afforded 120 mgm. of 11α,17α,21-trihydroxy 4-pregnene-3,20-dione (IV), m.p. 205–210°, $[\alpha]_D +$ 117° (methanol), $\lambda_{\mathrm{max.}}^{\mathrm{MeOH}}$, 242 mμ ($\varepsilon = 14,400$), $\lambda_{\mathrm{max}}^{\mathrm{KBr}}$ 3,442 (OH), 1,715 (20-CO), 1,653 and 1,615 (Δ^4-3-CO) cm.$^{-1}$. The 11,21-diacetate melts at 206–208°, $[\alpha]_D + 120°$ (methanol). The infra-red spectra of these compounds agreed with those of authentic samples.

From this experiment, 11α-hydroxylation of a steroid, which is one of the most important and common reactions among fungi, is carried out equally by Actinomycetes, fungi and bacteria. But, it may be worth mentioning that there is a remarkable difference in bioconversion patterns between Actinomycetes and fungi, for although both *Streptomyces* sp. and fungi, for example *Rhizopus arrhizus*, produced the same 6β, 11α-dihydroxyprogesterone as a dihydroxylated product from progesterone, they produced a different monohydroxylated product; *Streptomyces* sp. produced 6β-hydroxyprogesterone and, in contrast, *Rhizopus arrhizus* 11α-hydroxyprogesterone. So the enzyme systems of these two micro-organisms would appear to be different.

We wish to express our thanks to Prof. K. Tsuda and Prof. T. Asai of the Institute of Applied Microbiology, University of Tokyo, for their helpful advice and constant guidance throughout this work.

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[1] Eppstein, S. H., Meister, P. D., Murray, H. C., and Peterson, D. H., *Vitamins and Hormones*, **14**, 359 (1956). Visher, E., and Wettstein, A., "Adv. in Enzymol.", **20**, 237 (1958).

[2] Peterson, D. H., Murray, H. C., Eppstein, S. H., Reineke, L. M., Weintraub, A., Meister, P. D., and Leigh, H. M., *J. Amer. Chem. Soc.*, **74**, 5933 (1952). Fried, J., Thoma, R. W., Gerke, J. R., Herz, J. E., Donin, M. N., and Perlman, D., *J. Amer. Chem. Soc.*, **74**, 3962 (1952).

[3] McAleer, W. J., Jacob, T. A., Turnbull, L. B., Schoenewaldt, E. F., and Stoudt, T. H., *Arch. Biochem. Biophys.*, **73**, 127 (1958).

[4] Sugawara, S., Tsuruta, M., Shirasaka, M., and Nakamura, M., *Arch. Biochem. Biophys.*, **80**, 383 (1959).

[5] Colingsworth, D. R., Karnemoat, J. N., Hanson, F. R., Brunner, M. P., Mann, K. M., and Haines, W. J., *J. Biol. Chem.*, **203**, 807 (1953).

Histochemical Demonstration of a Species-Specific Trypsin-like Enzyme in Mast Cells

DURING the investigation of a new histochemical substrate, N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride, localization by a simultaneous azo coupling reaction of enzymatic activity to mast cells in human and dog tissues was noted. Recently, Benditt has described the presence of an enzyme in the mast cells of numerous species resembling bovine α-chymotrypsin. This enzyme hydrolyses the histochemical substrate 3-chloroacetyl-2-naphthoic acid anilide[1]. Although N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride has been shown in biochemical systems to be a substrate for trypsin and not for α-chymotrypsin, carboxypeptidase, liver esterase or 'leucine' aminopeptidase[2], it was thought of importance to attempt to define as closely as possible the characteristics of the enzyme in mast cells hydrolysing N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride and to differentiate it from other known mast cell enzymes.

N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride was synthesized by the mixed anhydride method[3]. Coupling of N^α-benzoyl-N^ω-nitro-*l*-arginine with β-naphthylamine was effected in 60 per cent yield, the product melting at 220–3°. The nitro group was removed by hydrogenation with a palladium catalyst in dilute hydrochloric acid to afford optically inactive N_α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride, m.p. 195–7° (ref. 4). Racemization apparently occurred in the course of activating the benzoylated amino-acid. 3-Chloroacetyl-2-naphthoic acid anilide was synthesized by the method of Gomori[5].

The incubation medium used in the histochemical experiments consisted of 30 mgm. of N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride and 10 mgm. of stabile diazotized 5-methyl-4-(nitro-*p*-tolylazo)-*o*-anisidine[6] (diazotized *o*-aminoazotoluene[7] may also be used) dissolved in 20 ml. of $M/20$ *p*H 7·0 *tris* buffer. Fresh frozen sections of skin or sections fixed for 16 hr. in neutral 10 per cent formalin at 4° C. were incubated in the above solution for 20–30 min., placed in a 1 per cent copper sulphate solution for 10 min., washed in running water, and mounted in glycerin gelatin containing several drops of a 1 per cent copper sulphate solution. Dark purple granules of the azo dye were observed only in mast cells (Fig. 1). The possibility of non-specific azo dye or naphthylamine binding by mast cell granules was eliminated when it was observed that tissue sections exposed to either a neutral aqueous suspension of the free naphthylamine (later coupled with diazonium salt) or the final azo dye showed no selective staining of mast cells. Enzymatic activity was completely abolished by di*iso*propylfluorophosphate (10^{-4} M), copper sulphate (2×10^{-3} M) and lead nitrate (2×10^{-3} M), but potassium cyanide (5×10^{-4} M and 10^{-2} M), sodium iodoacetate (10^{-3} M) and soybean trypsin inhibitor (1 mgm./ml.) failed to affect enzyme activity significantly. When stable tetrazotized di*ortho*anisidine[8] was used as the diazonium component in the histochemical system, inhibition of enzymatic activity was complete. This diazonium salt did not inhibit hydrolysis of 3-chloroacetyl-2-naphthoic acid anilide by mast cells.

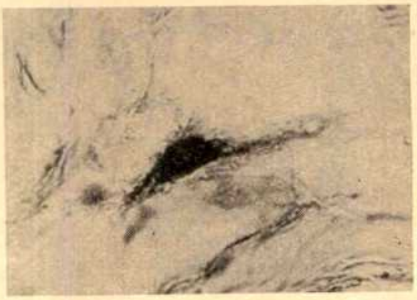

Fig. 1. Localization of trypsin-like enzyme in mast cell of human skin

Test-tube experiments with 0·5 ml./mgm. portions of salt-free crystalline α-chymotrypsin and trypsin (bovine) using N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride incorporated as substrate at a concentration of 5 mgm./ml. confirmed the failure of hydrolysis of N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride by chymotrypsin in 18 hr., and substantiated the rapid hydrolysis of the substrate by trypsin[2]. The method used for demonstration of hydrolysis in test-tube experiments was similar to that previously described[2] in which N^α-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride alone was incubated in the presence of enzyme and buffer. The released naphthylamine was estimated, following removal during incubation of aliquot samples, by azo coupling and extraction of the azo dye product. In a separate experiment the same stabilized diazonium salts used in the original histochemical procedure[6,7] were incorporated (0·5 mgm./ml.) in the trypsin incubating medium. No hydrolysis by trypsin as determined by extraction of azo dye was noted. This indicated that inhibition of trypsin activity by the diazonium salt or its stabilizer had occurred, though this inhibition did not occur in the histochemical system. Preliminary experiments to demonstrate competitive inhibition by the method of Benditt and Arase[9] revealed inhibition by N^α-benzoyl-*l*-arginine amide hydrochloride, a substrate rapidly hydrolysed by trypsin but not by chymotrypsin. Tissues of rat, rabbit, guinea pig, mouse, dog and human were examined for evidence of hydrolysis of N^{α}-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride by mast cells and only the latter two revealed enzymatic activity.

It is evident from the above results that the species-specific enzyme in mast cells demonstrated using N^{α}-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride as substrate has properties that relate it to, but do not identify it with, trypsin. The mast cell enzyme requires no apparent activation, is irreversibly inhibited by di*iso*propylfluorophosphate and competitively inhibited by N^{α}-benzoyl-*l*-arginine amide hydrochloride, but is not inhibited by soybean trypsin inhibitor or specific diazonium salts[6,7]. It differs from the enzyme described by Benditt in its apparent absence from rodent mast cells and its inhibition by a specific diazonium salt[8]. The competitive inhibition of N^{α}-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride by N^{α}-benzoyl-*l*-arginine amide hydrochloride is indicative, but not definitive evidence of its dissimilarity from the chymotrypsin-like enzyme. The fact that N^{α}-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride is not a substrate for α-chymotrypsin further substantiates this distinction. Inhibition of the trypsin-like enzyme by di*iso*propylfluorophosphate and lack of inhibition by cyanide distinguishes it from the aminopeptidase-like enzyme described in mast cells[10].

The physiological function of this trypsin-like enzyme in mast cells of certain species is not known, but it may possibly act to liberate secretory granule substance. It has been shown that lysis of mast cell granules is, in fact, proteolysis[11] and that activation of a trypsin-like enzyme (present in an inactive state in guinea pig mast cells) produces in these cells a hydrolysis of protein and release of histamine[12]. It has further been demonstrated that heparin is bound to protein at the time of its release from mast cells and that its release is dependent upon proteolysis[13]. It is therefore conceivable that as the result of a variety of injurious stimuli, a trypsin-like enzyme is either activated (rodents) or released from *in situ* inhibition (humans and dogs) within the mast cell cytoplasm. The incident intracytoplasmic proteolytic action of this enzyme may then liberate secretory granule substance[14].

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[1] Benditt, E., and Arase, M., *J. Exp. Med.*, **110**, 451 (1959).

[2] Riedel, A., and Wünsch, E., *Hoppe-Seylers Z. physiol. Chem.*, **316**, 61 (1959).

[3] Holland, G. F., and Cohen, L. A., *J. Amer. Chem. Soc.*, **80**, 3765 (1958).

[4] After this synthesis had been completed, preparation of Nα-benzoyl-*dl*-arginine-β-naphthylamide hydrochloride via the carbodiimide procedure was reported (cf. ref. 2).

[5] Gomori, G., *J. Histochem. Cytochem.*, **1**, 469 (1953).

[6] Fast corinth V from General Dyestuffs Co., New York.

[7] Fast garnet *GBC* from General Dyestuffs Co., New York.

[8] Fast blue *B* from General Dyestuffs Co., New York. Michrome blue 603 (diazotized 4′-amino-2′,5′-dimethoxybenzanilide), from Edward Gurr, Ltd., London, may also be used.

[9] Benditt, E., and Arase, M., *J. Histochem. Cytochem.*, **6**, 431 (1958).

[10] Braun-Falco, O., *Nature*, **183**, 51 (1959).

[11] Hill, M., *Nature*, **180**, 654 (1957).

[12] Ungar, G., and Damgaard, E., *J. Exp. Med.*, **101**, 1 (1955).

[13] Snellman, O., Sylvén, B., and Julén, C., *Biochim. Biophys. Acta*, **7**, 98 (1951).

[14] Keller, R., *Int. Arch. Allergy*, **11**, 328 (1957).

ANIMAL PHYSIOLOGY

Excretion of Rose Bengal in Bile

THE metabolism of rose bengal has been investigated by us in humans as well as in experimental conditions in animals. When we examined the dye by one-dimensional paper chromatography using 25 per cent ethanol and 5 per cent ammonia (1 : 1) as a solvent[1], we found it contained several components.

When a commercial preparation of rose bengal (Hopkins and Williams) was examined in ethanol–ammonia at least six components were distinguished in visible light, five red or orange-red (*R*1–*R*6) and one yellow (*Y*). The biggest spot was a red component (*R*3) with an R_F-value of about 0·50. Two other red components with R_F-values of 0·62 (*R*4) and 0·70 (*R*5) were second and third, respectively, in size. The other components appeared less prominent. In ultra-violet light all the above-mentioned components were seen fluorescing, the yellow spot having the brightest fluorescence. At least four faintly fluorescing spots (*F*1–*F*4), which could not be detected in visible light, were seen in ultra-violet light. In addition, radioactive preparations of rose bengal (Abbott) labelled with iodine-131 have been examined, and the distribution of radioactivity on the chromatogram estimated by scintillation counting and autoradiography. Some 51 per cent of the radioactivity was found localized to the spot *R*3. The *R*4 and *R*5 components contained 38 and 9 per cent respectively of the total radioactivity. We have found that there are great variations of the distribution of the radioactivity in the components *R*3–*R*5 of different batches of radioactive rose bengal, but the sum of radioactivity of *R*3–*R*5 usually amounts to more than 90 per cent of the total radioactivity. A slight blackening corresponding to the yellow spot was seen on the autoradiogram. Other details are seen in Fig. 1.

After intravenous injection into rats of the radioactive rose bengal together with a suitable amount of carrier, bile was collected and the excreted dye was extracted in ether after acidification of the bile. The ether-extracted rose bengal was subjected to paper chromatography in ethanol–ammonia. All components of the injected dye were recovered and

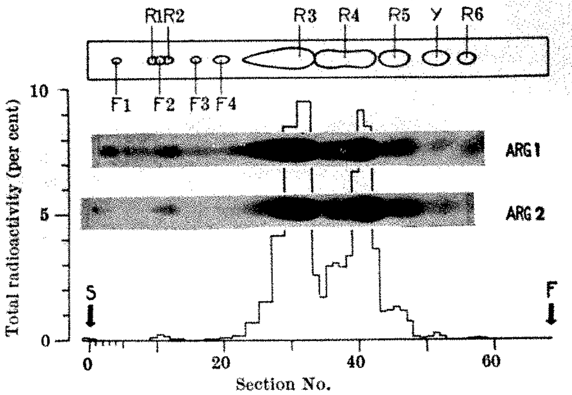

Fig. 1. Chromatography of rose bengal in ethanol–ammonia. *S*, starting point ; *F*, solvent front. *R*1–*R*6, red components, and *Y*, yellow component, seen in visible light ; *F*1–*F*4, fluorescing components only seen in ultra-violet light of the stable dye. *ARG* 1, autoradiogram of a chromatogram of a radioactive stock solution of rose bengal. *ARG* 2, autoradiogram of a chromatogram of radioactive rose bengal recovered from bile. The diagram shows the distribution of radioactivity on the chromatogram

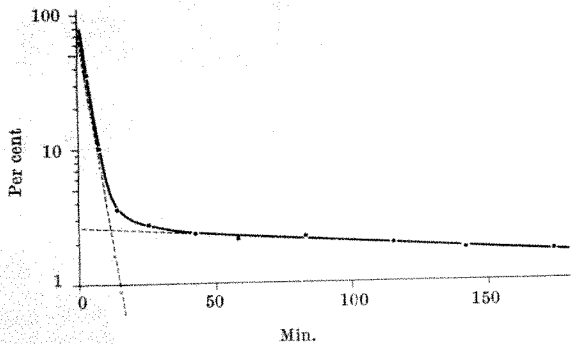

Fig. 2. Blood disappearance curve of radioactive rose bengal in the rat

in about the same proportions. Even the yellow component was excreted in the bile (Fig. 1).

The blood disappearance curve of rose bengal in the rat after injection of an iodine-131 labelled dye could be expressed as an approximate bi-exponential function. The disappearance rate of one phase was very slow; that of the other rather fast. When extrapolated to zero, the slow phase did not amount to more than 4 per cent of the dose injected (Fig. 2).

In accordance with Lima and Pieroni[2], we have found that rose bengal is not a pure compound but a mixture of several chemical substances. The explanation of the bi-exponential blood disappearance curve, however, can scarcely be based on this fact because the liver, as indicated above, is capable of removing all components of rose bengal from the blood. Furthermore, since the proportions between the components are roughly the same in the bile as in the injected material, no other organ seems to be responsible for a specific removal of any of the components. There is still a possibility that the liver might remove one of the small components of rose bengal from the blood at the slower rate. Thus we injected large amounts of radioactive rose bengal into rats. When the blood disappearance curve had reached its 'tail' portion the animals were killed and blood serum collected. When this radioactive serum was injected into rats the usual bi-exponential blood disappearance curve of rose bengal appeared.

The bi-exponential curve can be explained by assuming that there is a back-flow into the blood of a fraction of the amount of rose bengal in the liver cells. It then appears as if the percentage of the dye eliminated from blood will diminish in time and reach a low value.

We have observed that a back-flow to the blood of the rose bengal of the liver cells really can occur; for example, when rose bengal is injected 15 min. after bile obstruction the usual initial fall in the blood curve is followed by an increase in the blood radioactivity.

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[1] Ishida, Y., Ihagaki, N., Shiota, A., and Watanabe, R., *J. Pharm. Soc. Japan*, **73**, 736 (1953).

[2] Lima, F. W., and Pieroni, R. R., *Nature*, **184**, 1065 (1959).

Inhibitory Actions of γ-Aminobutyrylcholine

Since the existence of γ-aminobutyrylcholine in the mammalian brain and the possibility of its being a neurotransmitter have been reported[1], new findings on this substance have been reported by several investigators. Takahashi *et al.*[2] noticed its inhibitory effect on the electrical activity of the brain. McLennan[3] found that the second active ingredient of his factor 1 can be accounted for by this compound biologically but not chromatographically; and Kewitz[4] claims to have isolated γ-aminobutyrylcholine in crystalline form from the brain of the pig. In view of its anticholinergic and antihistaminic activities[1], we thought it worth while to study further its inhibitory properties on some other preparations.

The isolated heart of a clam, *Meretrix lusoria* Röding[4], reacted to γ-aminobutyrylcholine with decrease of tonus and the amplitude of beats. This action could be observed at a concentration of 10^{-2} *M* but not at 10^{-3} *M*. Washing removed all the effect. Similar action was exhibited by another choline ester of amino-acid like glycylcholine which was even more potent than γ-aminobutyrylcholine in this respect, and was active at the concentration of 10^{-4} *M* but not at lower concentrations. Unlike γ-aminobutyrylcholine, γ-aminobutyric acid caused short lasting tonic contraction of the heart at 10^{-1} *M*, but it was inactive at 10^{-2} *M*.

In the crayfish intestine, inhibitory potency of γ-aminobutyric acid and of γ-aminobutyrylcholine was about the same, both being effective up to 10^{-4} *M* but not effective at 10^{-5} *M*.

On the neuromuscular preparation of the crayfish claw[5], γ-aminobutyric acid and γ-aminobutyrylcholine elicited qualitatively similar action, inhibiting contraction of the indirectly stimulated closer muscle. After injection of either one, the height of its contraction showed a continuous decrease, but their effect was removed by washing with pure crayfish solution. γ-Aminobutyric acid was more potent; at 10^{-1}–10^{-3} *M* it caused complete block of the contraction and at 10^{-4} *M* the inhibitory action was still distinctly noticeable. γ-Aminobutyrylcholine also caused complete loss of contractility at 10^{-3} *M*, but not at lower concentrations, though its inhibitory effect was still noted at 10^{-4} *M*. Butyrylcholine was less potent than γ-aminobutyrylcholine in this inhibitory action, and glycylcholine was much less active.

Thus, there are some differences in potency and quality of their inhibitory actions, depending upon preparations used for study.

The most pronounced effect was observed on the cat's sciatic-gastrocnemius preparation of Brown. Following intra-arterial injection of 0·02 μgm. of γ-aminobutyrylcholine, the muscular contraction by indirect stimulation decreased in height rapidly, then disappeared, but resumed later (Fig. 1). With higher dose (20·0 μgm.), increase of the tonus set in. With

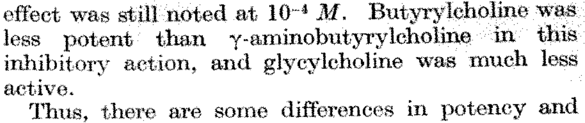

Fig. 1. Kymograph record of the contraction of cat's gastrocnemius muscle stimulated through the sciatic nerve with square wave stimulator. γ-Aminobutyrylcholine, 0·02 μgm. At the point indicated, it was injected intra-arterially in the dose of 0·02 μgm.

much higher dose (200·0 μgm.), the muscle reacted with irregular fibrillary twitches, instead of normal contraction, to the stimulation of the nerve, and after a while it became quite unresponsive. Such an extreme potency of γ-aminobutyrylcholine on the mammalian neuromuscular transmission is noteworthy. Besides, this property is dissimilar from that of the inhibitory factor of McLennan, which is reported not to affect this preparation[6]. Glycyl-, propionyl-, and butyryl-choline were also inactive at doses of 0·4, 4·0, and 40·0 μgm., except that 40·0 μgm. of propionylcholine caused temporary fall of the size of contraction.

Thus, γ-aminobutyrylcholine exerts a definite inhibition on various activities. A part of it may be due to its anticholinergic activity already reported[1]. However, since it has other properties such as antihistaminic action in addition, its inhibitory activity should not be ascribed solely to competition at the acetylcholine receptor. Further studies are necessary for understanding the mechanism of its inhibitory actions.

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[1] Kuriaki, K., Yakushiji, T., Noro, T., Shimizu, T., and Saji, Sh., *Nature*, **181**, 1336 (1958).

[2] Takahashi, H., Nagashima, A., and Koshino, Ch., *Nature*, **182**, 1443 (1958).

[3] McLennan, H., *J. Physiol.*, **146**, 258 (1959).

[4] Kewitz, H., *Naturwiss.*, **46**, 495 (1959).

[5] Yoshihara, H., and Kuriaki, K., *C.R. Soc. Biol.*, **151**, 1462 (1957).

[6] Florey, E., *Canad. J. Biochem. and Physiol.*, **34**, 669 (1956).

[7] Florey, E., and McLennan, H., *J. Physiol.*, **129**, 384 (1955).

Fracture Healing by Vitamin K

THE aim of this short communication is to report on the pronounced effect of vitamin K in speeding up the rate of bone healing. Some clinical observations (Bouckaert, A., and Hart Kliniek, H., personal communication) and preliminary experiments (Bouckaert, J. H., and Oyaert, W., personal communication) indicated that vitamin K had a real effect on all collagen tissues and especially bone tissue.

Adult male and female rabbits and rats were used in this study. Complete transverse fractures of the left tibia were produced by an open operation under ether anæsthesia. In the case of rabbits intramedullary pins were used to fix the fractured ends of the bone. In rats the fractures were supported by maintaining the fibulæ intact. The operation wound, which was thereafter closed with catgut, healed uneventfully in nearly all cases. All animals were kept under the same dietary conditions on a well-balanced ration, fed and watered *ad libitum*. Various groups of animals received vitamin K while others served as controls.

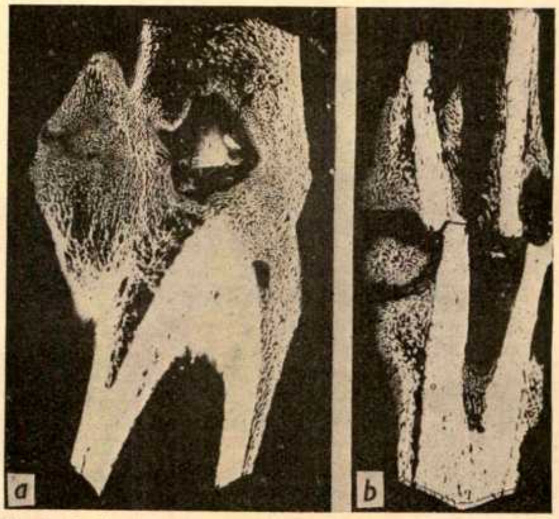

Fig. 1. Radiomicrographs from fractured tibiæ of *a*, an animal receiving vitamin K ; *b*, a control animal

Fig. 1*a* is a radiomicrograph of a fractured rabbit tibia after treatment with vitamin K. It shows a well-developed periosteal callus and the interfragmentary space woven with primary bone in a comparatively high degree of mineralization. Fig. 1*b* is a radiomicrograph of a control animal which received no medicaments. Here the periosteal callus is not complete. The space between the fractured ends is occupied with tissues devoid of any mineral deposition. Ordinary radiographs showed a complete continuity of the fractured bone of the treated animal in contrast to persistence of the fracture site in the control.

Full details about the further steps in our study with vitamin K singly or combined with other therapeutics as well as its exact mode of action will appear elsewhere.

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Effect of Fœtal Hypophysectomy on the Fœtal Liver Fat in the Rabbit

ANGEVINE[1], in his study of the pituitary and adrenals of 20 human anencephalic fœtuses, noted that in practically every case the subcutaneous pannus of fat was unusually thick especially over the shoulders and thorax. This observation has been confirmed in a number of full-term or near full-term anencephalics which are now being studied. Jost[2] has recently shown that fœtal thyroidectomy in the rabbit results in an increase in total fat (determined by ether–alcohol extraction), which does not occur if thyroxine is injected into the fœtus at the time of the thyroidectomy. This raises the possibility that the increase in fat in the human abnormality is due to a fœtal hormonal disturbance, and this view is supported by the knowledge that the pituitary of anencephalics is reduced in size and that secondary to this the adrenals are extremely small[1]. It is therefore possible that the hypofunction of the pituitary either directly, or indirectly by affecting other endocrine glands, determines the increase in fat found in the human anencephalic monster.

To study this aspect of anencephaly further it was decided to investigate the effect of removing the

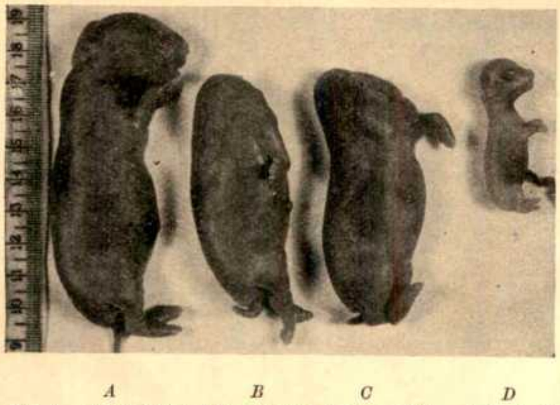

Fig. 1. Effect of fœtal decapitation in the rabbit. *A*, Control fœtus, 28 days old; *B* and *C*, fœtuses decapitated at 21 days and developed *in utero* to 28 days; *D*, a 21-day rabbit fœtus, showing the size of the fœtus at the time of the operation

fœtal pituitary in the rabbit on the total fat content of the fœtal liver.

Five rabbit fœtuses were decapitated (using the method described by Jost[3]) to remove the fœtal pituitary gland. The operation was performed between the twentieth and twenty-first day after mating, and the headless fœtuses were left within the uterus to continue growing. After a further seven to eight days of intrauterine development the fœtuses and the litter mates were removed by Cæsarian section, giving a total of five decapitated fœtuses and fifteen litter mate controls for study. The weights of the operated fœtuses were determined and compared with the headless weights of the control fœtuses.

The liver was dissected from all fœtuses and the fats extracted with ether and expressed as a percentage of the dry weight of the liver.

It was found that although the decapitated fœtuses were very slightly smaller by weight than the headless controls, this difference was not significant. Fig. 1 shows two 28-day fœtuses which were decapitated at 21 days, compared with a 28-day litter mate control; the small fœtus is 21 days old and shows the size at the time of decapitation.

Table 1 shows the results of the estimation of liver fat.

Table 1. TOTAL FAT OF LIVER OF DECAPITATED AND CONTROL RABBIT FŒTUSES

Fœtuses	Age at death	No.	Total liver fat (per cent of dry weight)	± *S.E.*
Controls	28–29 days	15	41·3*	± 1·5
Decapitated (at 20–21 days)	28–29 days	5	49·4*	± 2·3

* This difference is significant. $P < 0{\cdot}01$.

These results demonstrate that the operated rabbit fœtus had a significantly greater content of fat in the liver than the controls. This effect is likely to be due to the removal of the fœtal pituitary, and the work of Jost[2] suggests that the effect may be mediated secondarily through the fœtal thyroid.

These findings, together with those of Jost[2], suggest that the increase in fat noted by Angevine[1] in the human anencephalic fœtus and confirmed in this investigation is an endocrine effect resulting from the hypofunction of the fœtal pituitary and possibly other endocrine glands.

It is of interest in this connexion to note that a recent investigation[4] showed that the thyroids of two anencephalics showed the ability to concentrate iodine and make thyroxine.

I am indebted to Profs. E. W. Walls and A. Jost for help and advice in this investigation.

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[1] Angevine, D. M., *Arch. Path.*, **26**, 507 (1938).

[2] Jost, A., and Picon, L., *C.R. Acad. Sci., Paris*, **246**, 1281 (1958).

[3] Jost, A., *Arch. Anat. Micro. Morphol. Exp.*, **40**, 247 (1951).

[4] Yamazaki, E., Noguchi, A., and Slingerland, D. W., *J. Clin. Endocrinol. and Metabol.*, **19**, 1437 (1959).

A Neurohypophyseal Principle in the Elasmobranch Pituitary

THE elasmobranch pituitary gland consists of three distinct and largely separate regions (Fig. 1). Two of these, the rostral and ventral lobes, appear to represent the pars anterior of higher vertebrates, while the third, the so-called neurointermediate lobe, consists of intimately interdigitated neural and intermediate tissue[1]. Scharrer[2] has described a pre-optico-hypophyseal system in *Scyliorhinus stellare*, and our own unpublished histological studies on this system in *S. caniculus* have demonstrated the presence of material which is Gomori-positive, aldehyde–fuchsin-positive and stains by the performic acid–alcian blue technique of Adams and Sloper[3]. However, in spite of the undoubted existence of a neurohypophyseal system in elasmobranchs, evidence for the existence of the oxytocic and vasopressor principles characteristically present in the mammalian neurohypophysis is conflicting[4-8]. The investigation reported here attempts to resolve the discrepancy between the morphological and physiological findings.

Pituitary glands were removed from freshly killed *Squalus acanthias*, *Scyliorhinus caniculus*, *Raia clavata*, *R. batis* and *R. naevus*, separated into their constituent lobes, and dried in acetone. The resulting powders were extracted with 0·25 per cent acetic acid by the standard method for ox posterior lobe pituitary, and assayed against international standard posterior lobe pituitary powder, 'Pitocin', or 'Pitressin' (Parke Davis), for oxytocic activity, by the rat uterus and milk ejection methods of Holton[9] and Cross and Van Dyke[10] respectively, for antidiuretic activity[11], and for vasopressor activity[12]. By these means oxytocic, milk ejection and antidiuretic activities were found

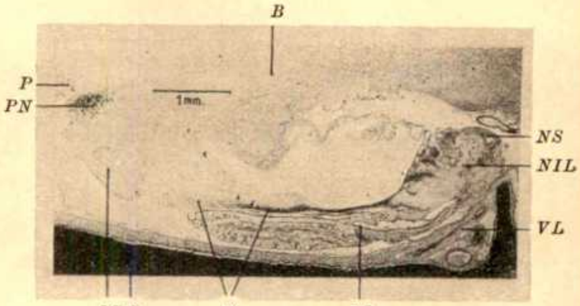

Fig. 1. *Scyliorhinus caniculus*. Parasagittal section of infundibular region of the brain. *B*, Brain tissue; *C*, cartilaginous floor of the skull; *H*, pre-optico-hypophyseal tract; *NIL*, neurointermediate lobe pituitary; *NS*, neurosecretory material in neurohypophyseal region of *NIL*; *OC*, optic chiasma; *P*, paraphysis; *PN*, preoptic nucleus; *RL*, rostral lobe pituitary; *VL*, ventral lobe pituitary

Table 1. OXYTOCIC AND ANTIDIURETIC ACTIVITIES OF ELASMOBRANCH NEUROINTERMEDIATE LOBE EXPRESSED AS M.U./MGM. ACETONE-DRIED POWDER

Species	Oxytocic activity (isolated rat uterus)	Oxytocic activity (rabbit milk ejection)	Antidiuretic activity (conscious rat)	Milk ejection oxytocic activity expressed as a percentage of rat uterus oxytocic activity	Antidiuretic activity expressed as a percentage of oxytocic activity	
					Rat uterus	Milk ejection
Squalus acanthias (pregnant ♀♀)	17·0	45	1·3	265	7·6	2·8
Scyliorhinus caniculus	13·2 ± 1·0	60	0·5	450	3·8	0·8
Raia clavata	17·0	60	0·13	353	0·7	0·2
R. batis	7·7 ± 0·5	35	0·6	454	7·7	1·7
R. naevus	10·0	—	0·5	—	5·0	—

to be present and were preferentially distributed in the neurointermediate lobe. No pressor effects were obtained from the amounts of material injected. Table 1 gives a brief summary of some representative results drawn from an extensive series of assays. It is evident from this that:

(1) The potencies, expressed in m.u./mgm. of dried powder, are low compared with international standard posterior lobe pituitary powder. These low figures are largely accounted for by the fact that elasmobranch neurointermediate lobe is composed mainly of intermedia tissue (Fig. 1).

(2) Very little antidiuretic activity is present in any of the elasmobranch powders. This confirms the earlier pioneer work of Heller[7] on dogfish and skate. Values are always below 9 per cent (average 5·1 per cent) of the rat uterus oxytocic figures, and below 2·8 per cent (average 1·2 per cent) of the corresponding milk ejection potency.

(3) The oxytocic activity of neurointermediate lobe powder is always approximately three times as high when measured by the rabbit milk ejection method as when measured by the rat uterus method.

The pharmacological properties of elasmobranch neurohypophyseal extracts have been studied in detail and have been shown to correspond closely to those of their mammalian counterparts. For example: All activities are stable in an acetone-dried powder, and as extracts in 0·25 per cent acetic acid; the oxytocic activity is unaffected by crystalline pepsin, crystalline trypsin, or atropine; the oxytocic and antidiuretic activities are destroyed by 0·01 *M* sodium thioglycollate at room temperature, by warming with 0·1 *N* sodium hydroxide, and by ultraviolet light; the oxytocic activity is destroyed by crystalline chymotrypsin. In 'four-point' rat uterus assays there is no significant deviation from parallelism between the assessed dose response curves for the elasmobranch agent and 'Pitocin'. In contrast to acetylcholine, and extracts of elasmobranch indifferent brain and hypothalamus, and in common with beef oxytocin, the elasmobranch neurointermediate lobe extract shows a long latency of action on the isolated rat uterus.

By definition, the ratios between milk ejection activity, rat uterus oxytocic activity and antidiuretic activity of international standard posterior lobe pituitary powder are 1 : 1 : 1. Analysis of all our results shows that these ratios in the elasmobranch material approximate to 3·5 : 1 : 0·05. A survey of the relevant literature demonstrates that this low titre of antidiuretic hormone relative to oxytocin is unique among the vertebrates so far examined. It seems unlikely that these discrepant ratios in elasmobranchs are due to contaminating inhibitors or activators since both 'Pitressin' and 'Pitocin', when added to elasmobranch neurointermediate lobe extracts inactivated by ultra-violet light, assay at their normal value, and there is the same high degree of destruction of all the activities by the various treatments described above. Recent work on oxytocin analogues has shown that single amino-acid substitutions alter the biological properties of the molecule, and analogues have already been synthesized which are more potent in releasing milk than in causing the rat uterus to contract when assayed against posterior lobe pituitary powder[13].

The antidiuretic activity of the elasmobranch materials examined is approximately 5·0 and 1·2 per cent of the corresponding values for oxytocic activity obtained by rat uterus and milk ejection assays respectively; these percentages approach the intrinsic antidiuretic activity of synthetic oxytocin. In the light of the low antidiuretic potency of the elasmobranch extracts and the discrepant oxytocic activity ratios, it is suggested that the neurointermediate lobe of the elasmobranch pituitary, so far as it is possible to generalize from five species, is exceptional in containing no discrete vasopressor/antidiuretic principle, though an oxytocic substance, not identical with beef oxytocin, is undoubtedly present. Recent work of Pickering and Heller[14], and Sawyer, Munsick and van Dyke[15], has demonstrated the presence of two oxytocin-like substances in the neurohypophyses of a variety of lower vertebrates (not elasmobranchs). Our range of assays is insufficient to say whether or not a similar situation obtains also in elasmobranchs, though if this is afterwards shown to be the case, it does not affect our submission that a modified type of oxytocin occurs in the elasmobranch neurohypophysis and that the vasopressor/antidiuretic hormone is absent.

This work, a detailed report of which will be published elsewhere, was supported by a generous grant from the Nuffield Foundation.

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[1] van de Kamer, J. C., and Verhagen, T. G., *Proc. Acad. Sci. Amst.*, C, **57**, 358 (1954).
[2] Scharrer, E., *Z. Zellforsch.*, **37**, 196 (1952).
[3] Adams, C. W. M., and Sloper, J. C., *J. Endocrinol.*, **13**, 221 (1956).
[4] Herring, P. T., *Quart. J. Exp. Physiol.*, **6**, 73 (1913).
[5] Herring, P. T., *Quart. J. Exp. Physiol.*, **8**, 245 (1914).
[6] Hogben, L. T., and de Beer, G. R., *Quart. J. Exp. Physiol.*, **15**, 163 (1925).
[7] Heller, H., *J. Physiol.*, **99**, 246 (1941).
[8] Waring, H., and Landgrebe, F. W., in "The Hormones", **2** (New York, 1950).
[9] Holton, P., *Brit. J. Pharmacol.*, **3**, 328 (1948).
[10] Cross, B. A., and Van Dyke, H. B., *J. Endocrinol.*, **9**, 232 (1953).
[11] Ginsburg, M., and Heller, H., *J. Endocrinol.*, **9**, 267 (1953).
[12] Landgrebe, F. W., Macaulay, M. H. I., and Waring, H., *Proc. Roy. Soc. Edin.*, B, **62**, 202 (1946).
[13] Berde, B., Doepfner, W., and Konzett, H., *Brit. J. Pharmacol.*, **12**, 209 (1957).
[14] Pickering, B. T., and Heller, H., *Nature*, **184**, 1463 (1959).
[15] Sawyer, W. H., Munsick, R. A., and van Dyke, H. B., *Nature*, **184** 1464 (1959).

HÆMATOLOGY

Paradoxical Effect of Magnesium Ions on Blood Coagulation

We have recently reported on the delay of whole-blood thrombin generation *in vitro* and *in vivo* which occurs when the concentration of magnesium is raised[1,2]. These results might have been thought to contradict the fact that addition of magnesium is known to enhance clotting of blood of which the calcium content has been lowered by previous passage through a suitable ion exchange column[3].

The inhibitory effect of magnesium on untreated blood has been explained on the basis of a competition between magnesium and calcium in processes where calcium is the more efficient catalyst[4]. We want to suggest that the present apparent contradiction can also be explained on similar lines because the outcome of the competition will depend on the concentration of either ion.

In decalcified plasma, magnesium alone will not restore coagulability. However, when sub-optimal amounts of calcium are added, magnesium will then enhance clotting. This can be understood if one assumes that some part or parts of the clotting mechanism can be activated by calcium alone, and that others can utilize magnesium though as a less efficient catalyst. Thus, if the amount of calcium is sub-optimal magnesium will be calcium-sparing; but if enough calcium is available to serve all clotting processes, added magnesium can then only displace the more efficient catalyst, and its effect will therefore be to retard clotting. Hence, the effect of a given concentration of magnesium on clotting will depend on the calcium concentration. Table 1 shows that with a calcium concentration of 5 m.equiv. per l., 10 m.equiv. per l. of magnesium is inhibitory, and with 2 m.equiv. per l. of calcium, 10 m.equiv. per l. of magnesium activates.

Table 1. Clotting Times (min.) of De-ionized Plasma diluted 10 : 1 with Additive

Ca⁺⁺ (m.equiv. per litre)	Mg⁺⁺ (m.equiv. per litre)			
	—	2	4	10
2	>60	11	12	32·5
3	15			
4	8·5			
5	7·5	7	8	11
6	7			
8	7			
10	7			
15	7	7	8	10
20	7·5			
30	9·5			
40	15			
50	>60			

Blood was taken into an ion exchange blood pack (Fenwall Laboratories, U.S.A.). Plasma was prepared by centrifuging at 1,500 r.p.m. for 20 min. 1 ml. of plasma added to 1 ml. additive in 8 × 75 ml. glass tubes incubated in a 37° C. water-bath. Clotting end-point was taken as the time when tubes could first be inverted without spilling (checked at ½-min. intervals).

As one might expect, with increasing quantities of magnesium the activating effect becomes less, and the inhibitory effect more pronounced. 2 m.equiv. per l. of magnesium will restore the clotting time of a calcium-poor plasma (2 m.equiv. calcium per l.) almost to normal, but will not inhibit the clotting of plasma containing normal amounts of calcium. On the other hand, 10 m.equiv. per l. of magnesium shows only little activation of the calcium-poor plasma, but marked inhibition at physiological levels. Furthermore, with the larger amount of magnesium the inhibition is diminished when calcium is present in high concentrations—bordering on those where calcium itself starts to become an inhibitor. This again illustrates the competition which exists between calcium and magnesium for clotting factors.

(This self-inhibitory effect of high calcium concentration is well-known, and has been explained[4] on a mechanical basis. The divalent calcium ion supposed to link two protein molecules will fail to form this bridge when it is in excess because too many of these molecules will have a calcium atom of their own.)

We wish to thank the Governors of St. Bartholomew's Hospital for a grant towards equipment.

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[1] Anstall, H. B., Huntsman, R. G., Lehmann, H., Hayward, G. W., and Weitzman, D., *Lancet*, i, 814 (1959).

[2] Huntsman, R. G., Hurn, B. A. L., and Lehmann, H., *J. Clin. Path.*, **13**, 88 (1960).

[3] Soulier, J.-P., Proc. Seventh European Congress of Hæmatology, 1959 (in the press).

[4] Greville, G. D., and Lehmann, H., *J. Physiol.*, **103**, 175 (1943).

Electrophoretic Patterns of Haptoglobins in Apes

The haptoglobin serum groups are Mendelian characteristics of normal people as are blood groups. Two common genes, Hp^1 and Hp^2, have been found and their frequencies in all white peoples studied have been about 0·4 and 0·6 respectively[1-7]. Gene Hp^2 seems to be much rarer in Negroes than in Whites[1,5].

The sera of most mammalian species seem to contain haptoglobin, but the only electrophoretic pattern that we have found in domestic and laboratory animals studied resembles closely that of Hp^1Hp^1 (Hp 1–1) people. On account of this observation we have studied blood samples of different primates listed below with the starch gel electrophoresis method described earlier[6].

Macacus rhesus	17 specimens
Macacus radiata	10 ,,
Papio hamadryas	1 ,,
Cercopithecus aethiops	1 ,,
Pan traglotydes	2 ,,

Sera of all apes studied gave the pattern *C* of Fig. 1, which closely resembles the pattern *D* (human Hp 1–1).

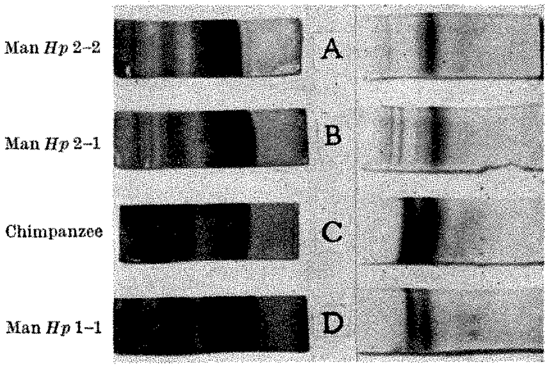

Fig. 1

It thus seems that if there is any haptoglobin polymorphism among *M. rhesus* or *M. radiata* demonstrable by starch gel electrophoresis, the gene giving *Hp* 1–1 like pattern is predominant.

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[1] Sutton, H. E., Neel, J. V., Binson, G., and Zuelzer, W. W., *Nature*, **178**, 1278 (1956).
[2] Laurell, A.-B., and Grubb, R., *Vox Sanguinis*, **2**, 312 (1957).
[3] Galatius-Jensen, F., *Acta Genet.*, **7**, 549 (1957).
[4] Fleischer, E., and Lundevall, J., Proc. Sixth Congr. Europ. Soc. Hæmatol., Pt. 2, 907 (1958).
[5] Allison, A. C., Blomberg, B. S., and Mrs. ap Rees, *Nature*, **180**, 824 (1958).
[6] Mäkelä, O., Eriksson, A. W., and Lehtovaara, R., *Acta Genet.*, **9**, 149 (1959).
[7] Moullec, J., and Fine, J. M., *Nature*, **184**, 196 (1959).

Serum Chromatography on Columns connected to a Common Source of Buffer Gradients

Recently there has been much progress in the separation of serum proteins by column chromatography. The best resolution of the protein components is theoretically obtained by the application of buffer and salt gradients to the column[1]. This principle has been applied during the elution of human serum from modified cellulose anion exchange columns when large numbers of chromatographic peaks were observed[2]. However, this technique is time-consuming, and it is difficult to produce an identical sequence of buffer gradients for each experiment. A modification using a single buffer gradient has been described for the separation of serum components of clinical interest[3], but this method reveals fewer components than the original technique. To overcome these difficulties a method with high resolving power for the simultaneous comparison of sera using the same system of buffer gradients has been developed and investigated.

A chromatographic comparison of human and animal serum on diethylaminoethyl cellulose under the same experimental conditions has not been reported. It was of interest during experiments with bovine serum to compare the results with the behaviour of human serum under identical conditions which were obtained by the simultaneous application to two columns of buffer gradients from the same source.

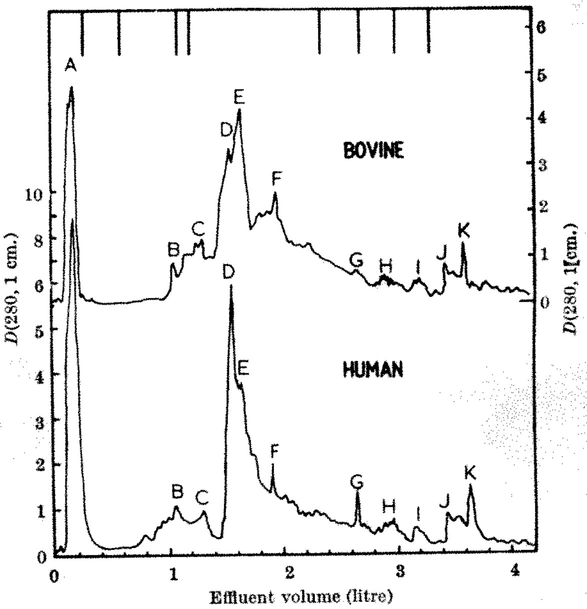

Fig. 1. Elution curves of two sera on diethylaminoethyl cellulose columns connected to the same buffer gradient source. Human serum (lower curve, left-hand ordinate) and bovine serum (upper curve, right-hand ordinate). The ordinates refer to the optical densities of a 1-cm. layer of the column effluent measured at 280 millimicrons wave-length (*D*(280, 1 cm.)). The positions of buffer changes are shown by the upper vertical lines. On both curves there are peaks emerging at similar effluent volumes. These corresponding peaks are labelled alphabetically in order of their elution

Experimental details were as follows: two columns (60 cm. × 2·5 cm.) were packed to a height of 25 cm. with diethylaminoethyl cellulose which contained 0·77 m.equiv. of ionizing groups per gm. They were mounted at different positions above an automatic fraction collector in a 4° C. room. The proteins of pooled bovine serum (100 ml.) and pooled human serum (100 ml.) were separated by gradient elution chromatography on separate columns essentially as in the original method[2], but common buffer gradients were generated for both columns in the same 2-l. reservoir connected to a 250-ml. conical flask with magnetic stirrer as mixing chamber. The common solution feed-tube was divided into separate inlet tubes for each column by a Y-shaped glass tube. Eluting solutions were forced through this system under regulated air pressure to give a flow through each column of about 20 ml./hr. Solutions used in the buffer reservoir were (*a*) 0·005 *M* sodium phosphate of *p*H 7; (*b*) 0·02 *M* sodium dihydrogen phosphate; (*c*) 0·03 *M* sodium dihydrogen phosphate; (*d*) 0·05 *M* sodium dihydrogen phosphate containing 0·02 *M* sodium chloride; (*e*) 0·05 *M* sodium dihydrogen phosphate containing 0·05 *M* sodium chloride; (*f*) 0·05 *M* sodium dihydrogen phosphate containing 0·1 *M* sodium chloride, and (*g*) 0·05 *M* sodium dihydrogen phosphate containing 0·5 *M* sodium chloride. Effluent fractions of about 10 ml. were collected. The volumes were accurately measured, and the ultra-violet absorption in a 1-cm. cell at 280 mμ was determined for each fraction.

Fig. 1 shows the elution curves of human and bovine serum. During the initial buffer gradient the sharp γ-globulin peak (*A*) emerged. It was followed by a large number of peaks which could be classified into 10 groups (*B*–*K*) each containing chromatographically distinct peaks.

Freeze-dried protein from each fraction was dissolved to give a 2 per cent solution which was examined by paper electrophoresis in 0·075 ionic strength veronal buffer of *p*H 8·6 followed by protein staining with bromphenol blue dye. As previously noted by Sober *et al.*[2], some difficulty was encountered in attempting to classify the results solely on the basis of mobilities. However, the patterns of the different groups showed the following distinct features: (*A*) protein only in the γ position, (*B*) protein in the β and γ regions, (*C*) protein in the γ position, (*D*) and (*E*) protein in the α and albumin regions, (*F*) major protein in the albumin region, (*G*) and (*H*) protein in the α, β and albumin positions, (*I*), (*J*) and (*K*) protein in the α and β regions. In addition, all the fractions showed some protein in the γ-globulin area of the paper strips. It was noticed that corresponding peaks had similar electrophoretic composition and emerged at the same effluent volumes from both sera.

This suggests the possible identity of the components from both sera and shows the reproducibility obtained on two columns by this method. The differences in the heights of corresponding peaks of the two sera probably reflect the known species differences in the electrophoretic composition of these sera.

The results suggest that this method may be useful for the comparison of different samples of animal sera. These sera are commercially important in the production of antibodies and as tissue-culture nutrients. Large-scale separation of these proteins on a battery of columns connected to the same buffer source may have advantages over single large-column operation, since the homogeneous packing, handling, regeneration, thermal and mechanical stability, and vertical space requirements may be better in the former case. Analytical applications for the comparison of pathological sera with a normal control serum may also be of interest, as modified cellulose columns have recently been described for the separation of myeloma proteins[3], rheumatoid factor[4], and lupus erythematosus factor[5] from human serum.

The present work is part of a project on the chromatography of high molecular weight substances. The help and support of the Director of the Commonwealth Serum Laboratories during this work is gratefully acknowledged.

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[1] Alm, R. S., Williams, R. J. P., and Tiselius, A., *Acta Chem. Scand.*, **6**, 826 (1952).

[2] Sober, H. A., Gutter, F. J., Wyckoff, M. M., and Peterson, E. A., *J. Amer. Chem. Soc.*, **78**, 756 (1956).

[3] Fahey, J. L., McCoy, P. F., and Goulian, M., *J. Clin. Invest.*, **37**, 272 (1958).

[4] Williams, R. R., Stewart, L. C., and Jenkins, J. C., *Proc. Soc. Exp. Biol. Med.*, **99**, 554 (1958).

[5] Wilkens, R. F., Dreschler, M., and Larson, D. L., *Proc. Soc. Exp. Biol. Med.*, **99**, 645 (1958).

Decrease of the Copper Bands of *Helix pomatia* Hæmocyanin

A study of the absorption spectra of α- and β-hæmocyanin of *Helix pomatia* indicated that the copper bands of α-hæmocyanin, with their maxima at 346 and 580 mμ, are not quite reproducible[1]. As a continuous decrease of the copper-bands was liable to occur, the extinction of hæmocyanin preparations has been followed over several months.

The blood of several snails was collected and centrifuged during 20 min. at 10,000 r.p.m. in a water-cooled Pirouette (Phywe, Göttingen, Germany). Samples were treated as quickly as possible in various ways and stored at 4° under toluene in well-stoppered flasks. Care was taken to ensure full saturation with toluene during the whole procedure. The extinctions were measured in a Beckman *DU* spectrophotometer at 346 and 278 mμ on aliquots, immediately after dilution with a saturated borax buffer of *p*H 9·18. At this *p*H the contribution of light scattering to the total extinction is practically negligible[2].

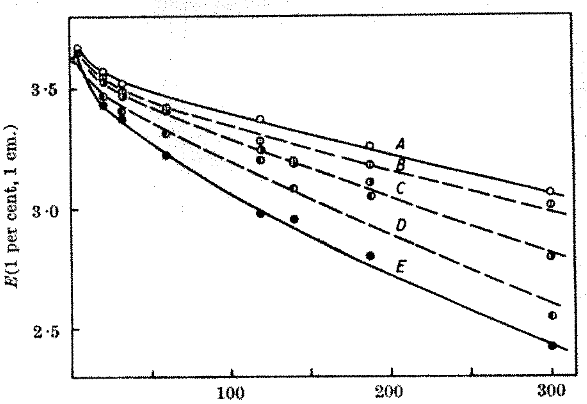

Fig. 1. The decrease during storage of the specific extinction coefficient of *Helix pomatia* hæmocyanin at 346 mμ

The specific extinction coefficients were calculated for the copper band at 346 mμ as a function of *E*(1 per cent, 1 cm.) = 14·16 at 278 mμ. As Fig. 1 shows, the extinctions gradually fall off, starting from the same value. The decrease is most pronounced for blood precipitated three times with ammonium sulphate (*p*H 5·3). The precipitate was dissolved in 0·1 *M* sodium acetate buffer, *p*H 5·7, after the first and the second precipitation. The third time it was stored with the supernatant solution (*E*). The drop of the extinction is smaller when this triple precipitation is followed by dialysis during one night against water at 4°, the precipitate having been dissolved in 0·1 *M* NaOAc, *p*H 5·7 (*C*). The protection is rather good after filtration of 100 ml. of blood on 10 gm. of activated charcoal ('Norit') (*B*) and very small after extraction with 1 volume of chloroform (*D*). Two ultracentrifugations (Spinco Model *L*, rotor No. 30, 2½ hr., 4°, 78,410*g*), followed by dialysis during 48 hr. against water (*A*), yielded good protection, comparable to the charcoal treatment (*B*).

The general shape of the copper bands remains unchanged, as indicated by a comparison of freshly prepared blood with some samples after 300 days storage. It is to be noted further that the extinction coefficients at 580 mμ follow exactly the corresponding values at 346 mμ (Table 1).

Table 1. RELATIONSHIP BETWEEN THE SPECIFIC EXTINCTION COEFFICIENTS OF THE COPPER BANDS AFTER 300 DAYS STORAGE (DILUTION WITH A SATURATED BORAX BUFFER, *p*H 9·18, 20° C.)

Treatment	*E*(1 per cent, 1 cm.) 346 mμ	*E*(1 per cent, 1 cm.) 580 mμ	E_{346}/E_{580}
D	2·55	0·154	16·5
C	2·79	0·171	16·4
B	3·01	0·179	16·8
A	3·06	0·185	16·5

In order to see if α- and β-hæmocyanin behave in a similar way, preparations of total hæmocyanin, made from blood collected during different years and stored under toluene at 4° as an ammonium sulphate precipitate for varying periods, were separated into the two proteins. Extinction coefficients at 346 mμ, determined at *p*H 9·18, are given in Table 2. For β-hæmocyanin four values out of five are very near 2·89. A high value of 3·30 was obtained on a β-preparation, prepared as quickly as possible from fresh blood. The extinction coefficients of some α-hæmocyanin preparations, stored under similar conditions, range from 1·78 to 2·61.

Table 2 SPECIFIC EXTINCTION COEFFICIENTS OF THE COPPER BAND AT 346 Mμ (PURIFIED PREPARATIONS, DILUTED WITH A SATURATED BORAX BUFFER, *p*H 9·18, 20° C.)

Preparation	Year of collection	Protein concentration (per cent (w/v))	*E*(1 per cent, 1 cm.)
β-Hæmocyanin	1952	0·1060	2·98
	1956	0·321	2·88
	1957	0·0563	2·90
(very fresh)	1957	0·0399	3·30
	1959	0·281	2·79
α-Hæmocyanin	1952	0·240	1·78
	1956	0·397	2·20
	1956	0·334	2·58
	1957	0·100	2·60
	1957	0·202	2·07
	1958	0·236	2·61
	1959	0·194	2·21
	1959	0·156	2·10

These observations clearly establish the parallel decrease of the copper bands of *Helix pomatia* hæmocyanin under ordinary storage conditions, β-hæmocyanin changing more slowly than α-hæmocyanin. To explain the decrease a bacterial action may well be ruled out, as the effect is highest in the presence of half-saturated ammonium sulphate. An organic substance, possibly aromatic, which is salted out by ammonium sulphate, could be responsible for this spectral decrease. Evidence has indeed been given for the presence of phenolic amines, for example tyramine, in the blood of some cephalopods[3].

Related observations have been made on the oxygen binding of hæmocyanins. A preparation of *Limulus polyphemus*, stored for several months under half-saturated ammonium sulphate, showed a 30 per cent decrease of the absorption band in the visible, which was accompanied by a 26 per cent lowering of the oxygen-binding capacity[4]. The hæmocyanin of *Helix pomatia* has further been reported to show during storage at − 10° a change from the sigmoid oxygenation curve to a rectangular hyperbola[5].

The ratio, however, of α- and β-hæmocyanin was completely normal in the preparations *A* to *E* after 300 days storage, as determined by dissociation tests in a Spinco ultracentrifuge model *E* and *M* sodium chloride at *p*H 5·8.

The antigenicity moreover of some hæmocyanin solutions, kept under toluene at 4°, did not undergo any noticeable change after ten years[6].

In conclusion it may well be stressed that the usual way of storing hæmocyanins as an ammonium sulphate precipitate, without any other treatment, proves to be the least satisfactory, if properties connected with the oxygen-binding sites are to be studied.

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[1] Heirwegh, K., Borginon, H., and Lontie, R., Fourth Int. Congr. Biochem., Vienna, 2 (1958).

[2] Lontie, R., Dessent, M., and Heirwegh, K., *Meded. vlaamse Acad. Kl. Wet.*, **13**, No. 1 (1951).

[3] Bacq, Z. M., and Ghiretti, F., *Arch. Int. Physiol.*, **59**, 288 (1951).

[4] Redfield, A. C., *Biol. Bull., Woods Hole*, **58**, 238 (1930).

[5] Horsmans, J., after Lontie, R., *Meded. vlaamse chem. Ver.*, **16**, 110 (1954).

[6] Litt, M., and Boyd, W. C., *Nature*, **181**, 1075 (1958).

PLANT PHYSIOLOGY

Vitamin E and Flowering of *Fragaria vesca* L. var. *semperflorens* Duch.

THE development of *Fragaria vesca* L. var. *semperflorens* Duch. is very sensitive to day-length[1]. In short-day conditions the plants do not reach the reproductive stage. Days more than 12 hr. long are required. Short-day treatment also inhibits flower formation of *Fragaria vesca* plants which have previously reached the flowering stage in long-day conditions. During a season in the open field, the number of flowers found on such flowering plants varies according to the day-length as shown in Fig. 1. A maximum of flowers is found in the inflorescences growing during August–October, which have been initiated during June–July, in the longest days.

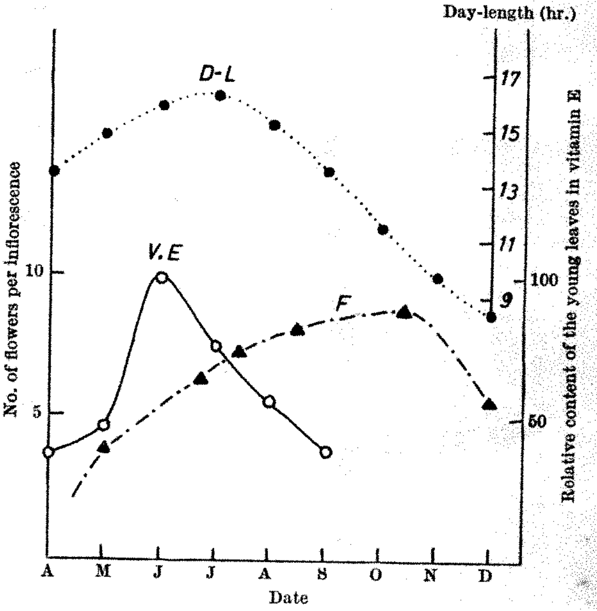

Fig. 1. Relations between day-length, number of flowers per inflorescence and vitamin E content of the young leaves in flowering plants of *Fragaria vesca* L. var. *semperflorens* Duch. (for explanations, see the text; *D-L*, day-length; *V.E.*, vitamin E content; *F*, number of flowers per inflorescence)

On the other hand, in some experimental conditions, runner plants of *Fragaria vesca* have been shown to improve their flowering capacity when vitamin E is applied externally to the leaves in low concentration[1]. It seemed, therefore, interesting to investigate the two following points: (1) Is the vitamin E content of *Fragaria vesca* plants grown in 8-hr. day (remaining vegetative) different from that of plants grown in 16-hr. day (beginning to form flowers)? (2) Is the vitamin E content of flowering plants related in some way to their flowering intensity?

Vitamin E was measured by the method of Meunier and Vinet[2] in the unsaponifiable fraction of the leaves extracted as described by Sironval[1]. The separation of the vitamin was obtained on 'Floridin *XS*' prepared following Kjölhede's method[3].

Table 1, measurements 1–3, answers the first question. The measurements were made in adult leaves of *Fragaria vesca* plants grown in 8-hr. days (remaining vegetative) or in 16-hr. days (beginning to form their first flowers). The temperature was maintained constant (20° C.) for measurement 1. Measure-

Table 1. Effect of Day-Length on the Vitamin E Content of the Adult Leaves of *Fragaria vesca* L. var. *semperflorens* Duch. (per γ in 100 mgm. of the Unsaponifiable Fraction)

Measurement No.	Temperature	8-hr. day	16-hr. day
1	20° C. constant	219	430
2	variable	255	490
3	variable	202	480
4	variable	185*	420

* This series has first been grown in long-days. It was transferred to short-days at the flowering stage. The measurement is done 2 months after the transfer (see text).

ments 2 and 3 were made on plants grown in the open field. It appears from Table 1 that the content of vitamin E per 100 mgm. of the unsaponifiable fraction is regularly about twice as high in long-days than in short-days. Calculated on a dry-weight basis, the recorded difference is still more marked.

Measurement 4 of Table 1 is concerned with the second question. Flowering plants grown in 16-hr. day were transferred to 8-hr. day. They were compared to the control remaining in 16-hr. day. After the transfer to short-day the flowering intensity strongly diminished, in accordance with previous observations[1]. Within two months after the transfer, the vitamin E content of the adult leaves of the transferred plants dropped to about half, while the content of the control remained constant.

Another observation concerned with the second question is recorded in Fig. 1, which gives for flowering plants in the field the normal variations of the vitamin E content of the young leaves near the apex (4 cm. high) during the year 1958. These variations are compared with the variations of the flowering intensity and those of day-length. It is seen that the vitamin E content is first low at the beginning of the spring; it rises rapidly until June, and falls after that. The maximum in June corresponds approximately to the maximum of day-length. It appears just before the maximum rate of flower initiation, at the moment of the rapid increase of the number of flowers per inflorescence. This has been found also during the year 1957 (Table 2). However, as shown in Table 2, the seasonal variations of the content in vitamin E is less evident in adult and old leaves than in young ones. The content drops with the ageing of the leaves, as we see when considering the underlined figures of Table 2, which are concerned approximately with the same set of ageing leaves.

It is concluded that the vitamin E content of *Fragaria vesca* leaves is very sensitive to day-length, and that this feature is related in some way to the action of day-length on flower formation. Normal flower formation seems to require a high vitamin E content in the leaves. This is seen not only in experiments with extreme day-length (8-hr. and 16-hr.), but also in ordinary culture in the field. In this last case, the occurrence of a high content in vitamin E in the young leaves surrounding the apex, at the moment of the active increase of flower initiation in June, strongly supports the idea that the vitamin plays a part in the promotion of flowering.

Table 2. Variations of the Vitamin E Content of the Leaves of Flowering *Fragaria vesca* L. cultivated in the Field during the Years 1957 and 1958 (100 for 1957, 62γ per 100 mgm. dry weight; for 1958, 71γ per 100 mgm. dry weight)

Time of observation	Young leaves		Adult, just expanded leaves		Old leaves	
	1957	1958	1957	1958	1957	1958
April	39	37	—	—	—	—
May	28	45	21	27	30	10
June	100	100	25	44	10	38
July	26	73	42	53	12	32
August	18	56	16	38	21	45
September	—	38	26	23	14	11

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[1] Sironval, C., *C.R. Rech. IRSIA, Bruxelles*, **18**, 1 (1957).

[2] Meunier, E., and Vinet, A., *C.R. Acad. Sci., Paris*, **211**, 611 (1940).

[3] Kjölhede, K., *Z. Vitaminforsch.*, **12**, 138 (1942).

Chlorogenic Acid of Lettuce Seeds

The high concentration of chlorogenic acid in lettuce seed has not previously been reported. Fig. 1 compares the ultra-violet absorption spectrum of a methanol solution of chlorogenic acid, 0·02 gm./l., with that of a methanol extract of 1 gm. of lettuce seed made up to 1 litre. The lettuce seed had been extracted continuously with methanol for 16 hr. in a Soxhlet apparatus. Paper chromatography of the extract showed the predominant fluorescent component to be chlorogenic acid. Several other fluorescent components which after elution show the same ultra-violet absorption spectra as chlorogenic acid are also present on the chromatograms. These are probably isomers of chlorogenic acid, but could be other depsides of caffeic acid. Caffeic acid itself is not a major component. Assuming that the compounds which have the same absorption spectrum as chlorogenic acid are isomers and have the same molecular extinction coefficient at 330 mμ, 2 per cent of the weight of the dry seed is chlorogenic acid and its isomers.

In order to investigate the possible role of this large amount of chlorogenic acid, lettuce seeds were germinated for as long as two weeks and the seedlings from 1 gm. of seeds were extracted with methanol.

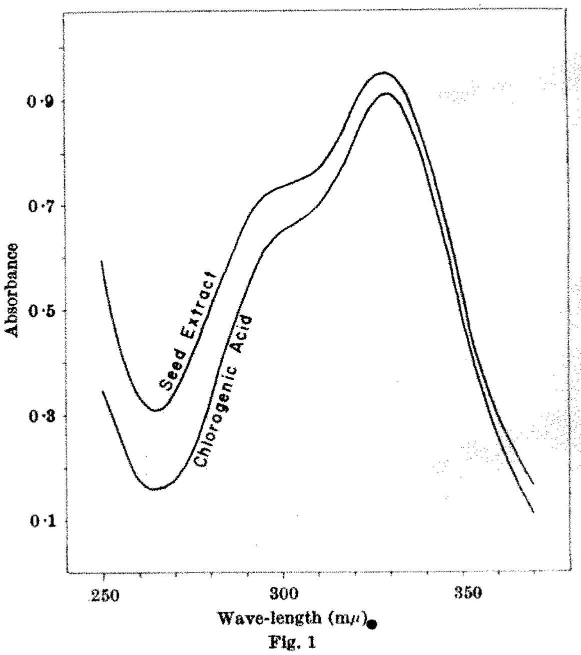

Fig. 1

The total amount of chlorogenic acid did not change during germination. By separating the cotyledons from the seedlings it was shown that the chlorogenic acid remained in the cotyledons of the seedlings with very little, if any, being translocated into the stem or roots. The possibility that chlorogenic acid was actively turning over cannot be ruled out. However, the apparent lack of utilization suggests that chlorogenic acid in lettuce seed may be an end-product which accumulates to a high concentration in the seed, but serves no metabolic function.

The lettuce seed also contains an active polyphenol oxidase as Mayer[1] has shown. Thus, the chlorogenic acid exists in the seed along with a polyphenol oxidase without interacting even in the wet, imbibed seed where metabolic processes are occurring. Mason[2] has suggested theoretically that polyphenol oxidases act as hydroxylating enzymes *in vivo* and assume their oxidase activity only when the cell is broken. According to this view the polyphenol oxidase which destroys the chlorogenic acid when the cell is broken might be involved in its formation and accumulation in the seed.

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[1] Mayer, A. M., *Enzymologia*, **16**, 277 (1954).

[2] Mason, H. S., *Nature*, **177**, 79 (1956).

Nitrogen Feeding of Apple Trees by Post-Harvest Urea Sprays

In the early post-bloom period it is common to spray apple trees with a 0·5 per cent urea solution which, when repeated several times, results in a clear increase of leaf chlorophyll and leaf total nitrogen in comparison with leaves of unsprayed trees[1]. Higher concentrations may cause leaf injury. It seems unlikely that this injury caused in the late autumn would reduce to any extent the total amount photosynthesized during the passing season. This would permit the use of comparatively concentrated spray solutions at that time. The absorption of nitrogen may be proportional to the concentration of the urea solution[2].

Prior to abscission some 40–50 per cent of the total nitrogen of the leaves is re-absorbed by the tree[3]. This nitrogen makes up the whole, or a part, of the nitrogenous reserves which are utilized the following growing season. Apart from securing the normal nitrogen needs of the tree, a large reserve may effect early starting of growth in the spring in comparison with trees of low reserves[4].

As is shown in Fig. 1, a spray in the middle of October with a 4 per cent urea solution has increased the total organic nitrogen content of the leaves by 51 per cent within two days after application. When attention was first directed to urea sprays as a means of supplying apple trees with nitrogen, the registered increase in leaf nitrogen was approximately 15 per cent after four applications[5]. Fig. 1 shows further that absorbed nitrogen has migrated from the leaves into the tree. By the time of leaf fall (November 12) the sprayed trees had spurs, with developed blossom buds, containing 31 per cent more nitrogen than spurs of unsprayed trees. The corresponding figure for terminal shoots is 16 per cent.

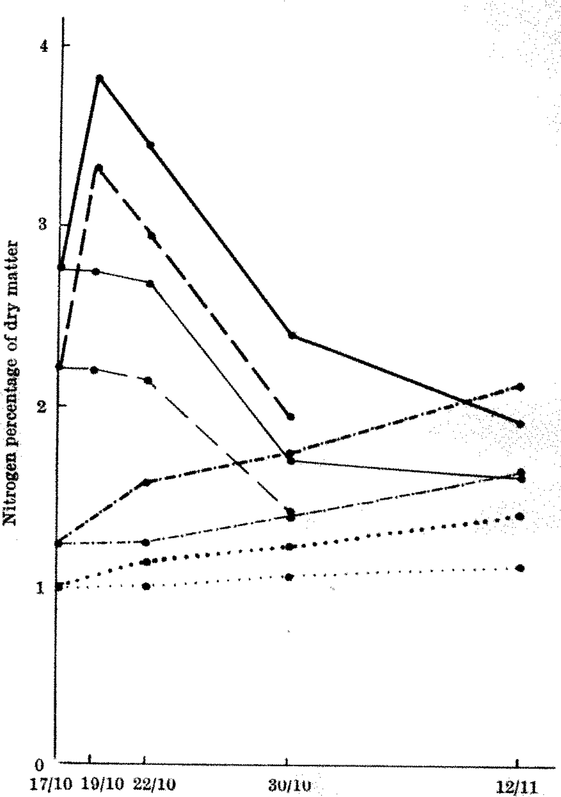

Fig. 1. Nitrogen content of different parts of apple trees sprayed with a 4 per cent urea solution on October 17 (heavy lines), and unsprayed control trees (thin lines). Shoot leaves, ———; spur leaves, — — —; shoots, ...; and spurs, -·-·-·-. All plant parts were washed thoroughly before drying and analysis

The leaf injury caused by the 4 per cent urea spray was some marginal browning of spur leaves and basal shoot leaves, while leaves at the tips of the shoots appeared unaltered.

It is suggested that the amount of nitrogen absorbed by the trees from a single spray might compare to a net intake of 30–40 kgm. nitrogen per hectare. (The amount of leaves in an established orchard is assumed to be of the order of magnitude of 3,000 kgm. dry weight per hectare.) This amount of nitrogen is well above what the trees may be expected to make profitable use of, besides the nitrogen supplied by a normal organic matter metabolism of the soil.

We lack knowledge concerning the responses of apple trees to nitrogen fertilization[6], which is at least partly due to the sources of errors in the experiments carried out. The experience gained during the past autumn, in experiments with the suggested method of nitrogen feeding, seems to promise that accurately known amounts of nitrogen may be introduced into the trees by post-harvest urea sprays. As large effects can be obtained, it would serve profitably in studies on the importance of nitrogenous reserves, as well as in studies on nitrogen metabolism.

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[1] Boynton, D., "Ann. Rev. Plant Physiol.", **5**, 31 (1954).

[2] Boynton, D., *et al.*, *Proc. Amer. Soc. Hort. Sci.*, **62**, 135 (1953).

[3] Murneek, A. E., *Proc. Amer. Soc. Hort. Sci.*, **27**, 228 (1930).

[4] Oland, K., *Physiol. Plant.*, **12**, 594 (1959).

[5] Hamilton, J. M., *et al.*, *Proc. Amer. Soc. Hort. Sci.*, **42**, 123 (1943).

[6] East Malling Res. Sta., Ann. Rep. (1959).

RADIOBIOLOGY

Reduction of the Radioactive Content of Milk by a Feeding Procedure

A METHOD for reducing the fall-out content of milk has been investigated. This is a preliminary report on the results obtained in a short-term study in this area. There are to-day commercially available 'grass incubators' which hydroponically germinate various grains to be used as a substitute for fresh pasturage. It is claimed that the product of these incubators fed to cattle on dry lot maintains the cows in a high production of superior-quality milk to an extent which makes the purchase of such incubators economically desirable. The increased use of these feeding systems in this area raised the question to us concerning the effect this substituted diet may have on the radioactive content of milk.

In the experimental work a total of twenty dairy cows was used, all of which had been under the same conditions for several weeks prior to the experiment. The cows were divided into two groups of ten animals each; the control group was kept on natural pasture during the experiment while the experimental group was confined in dry lot and had only incubator-germinated oats as their source of green pasturage. These oats were allowed to germinate for 6 days in the incubators under hydroponic culture and then were fed as intact mats of seed and shoot to the cows. Cultivation was according to the manufacturer's directions (Buckeye Incubator Co., Springfield, Ohio). Each cow in the experimental group was fed 20 lb. of incubator feed daily. Both groups were fed their usual hay and grain diet. Equal aliquots of milk were taken from each cow at each milking and pooled into a weekly sample for each group. Thus the analyses for radioactive content were made on weekly aliquots of pooled samples from each of the two groups. The total volume of each sample was measured and a 4-l. aliquot evaporated to 2 l. in a sand-bath. The residual liquid was counted for cæsium-137 and potassium-40 content. The counting was performed with an 8 in. × 4 in. sodium iodide (Tl) crystal feeding into a 256 channel analyser (Vanderbilt University Low Level Whole Body Counting Facility supported by: United States Atomic Energy Commission Grant No. *AT* (40-1)-240 and Contract No. *DA*-49-007-*MD*-995 with the Army Medical Research and Development Command). The samples were counted for 1 hr., which made possible net counts for cæsium-137 and potassium-40 in the range 500-3,000 counts. The results are given in Table 1 as net counts which are the gross counts less background. Background was run on a solution of 12·5 gm. potassium chloride in 2,200 ml. distilled water, to approximate the potassium content of milk.

The cæsium-137 counts in all of the seven weekly groups were significantly lower (more than twice the standard error of the difference) than the controls, with an average reduction of 38 per cent. The potassium-40 counts were significantly different only in the first and third weekly samples and fail to show a consistent trend.

The differences in the cæsium-137 counts between the control and the experimental groups becomes more significant when we consider that the substituted incubator feed comprises only a part of the total diet. A more detailed study is proposed.

We are indebted to Mr. George Ivey, of Little River Farms, Alpharetta, Georgia, for supplying the cattle and dairy facilities for this pilot study. Also, we wish to express our appreciation to Dr. George R. Menelly and Dr. Robert T. Lagermann for use of the 'Low Level Whole Body Counting Facility' at Vanderbilt University and to Mr. William Van Willis for his technical assistance.

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Table 1

Sample (weeks)	Cæsium-137 counts				Potassium-40 counts			
	Experimental	Control	Difference	Change (per cent)	Experimental	Control	Difference	Change (per cent)
0*	721 ± 99	732 ± 99	11 ± 140	1·5 ± 19	1,881 ± 84	1,868 ± 84	−13 ± 117	−0·7 ± 6
1	375 ± 97	930 ± 100	555 ± 139	60 ± 15	1,719 ± 83	2,175 ± 85	456 ± 119	21 ± 6
2	377 ± 97	713 ± 99	336 ± 139	47 ± 20	1,640 ± 82	1,664 ± 82	24 ± 116	1 ± 7
3	735 ± 99	1,060 ± 101	325 ± 141	31 ± 13	2,365 ± 86	2,059 ± 84	−306 ± 121	−13 ± 5
4	778 ± 99	1,158 ± 101	380 ± 141	33 ± 12	1,579 ± 80	1,569 ± 80	−10 ± 112	−0·8 ± 7
5	1,069 ± 101	1,667 ± 108	598 ± 147	36 ± 9	2,155 ± 86	1,841 ± 84	−314 ± 119	−15 ± 6
6	1,052 ± 100	1,499 ± 105	447 ± 145	30 ± 10	1,770 ± 83	1,995 ± 85	225 ± 118	11 ± 6
7	818 ± 99	1,117 ± 103	299 ± 143	27 ± 13	1,951 ± 85	1,811 ± 89	−140 ± 118	−7 ± 6
Average 1-7	743 ± 105	1,163 ± 111	420 ± 154	38 ± 14	1,883 ± 89	1,873 ± 89	−10 ± 126	−0·4 ± 7

* This sample was taken before the experimental animals were removed from pasture grass. These values were not included in the calculation of averages.

Radiosensitivity of the Pre-irradiated Yoshida Sarcoma

ACQUIRED radioresistance of cancer has been discussed clinically and experimentally by several authors[1-5]. The various opinions concerning its mechanism may be summarized as follows: (1) selection of cancer cells; (2) induction of mutation and its adaptation; (3) connective tissue response or tumour bed response. Experimental confirmations are less pronounced than clinical observations. One[1,2] of the recent experiments showed development of radioresistance using repeated frequent X-ray irradiations of intramuscular implantation of Ehrlich carcinoma, whereas others[3] noticed no change of radiosensitivity following frequent whole-body γ-ray irradiations of mice bearing Ehrlich ascites carcinoma. The results of the latter may be due to insufficient dose to develop a radioresistance.

It seemed, therefore, of interest to conduct an experimental study of radiosensitivity on the hypothesis that the most suitable way to induce radioresistance is to expose tumour cells to such a dose of irradiation that only a few cells could survive. By repeating this procedure to successive generations radioresistance might be obtained.

The experiment was initiated in 1955 using the Yoshida sarcoma. Mixed-bred rats weighing about 100 gm. were used. For the first transfer generation, 5×10^6 tumour cells were transplanted into the peritoneal cavity, and 4 days later this tumour-bearing rat received a whole-body X-irradiation of 1,000 r. Physical factors were 185 kVp., 6 m.amp., half-value layer 0·76 mm. of copper, focus-peritoneal cavity distance 23 cm. and dose-rate 74·1 r./min. in air. 1 hr. later, the same number of tumour cells was transplanted from this rat into the peritoneal cavity of the other rats to make the 2nd generation. After the proliferation of tumour cells at the 2nd generation had occurred in the peritoneal cavity, whole-body irradiation was repeated. 1 hr. later the same number of tumour cells from the 2nd generation rat was transplanted intraperitoneally, making the 3rd generation. Thus, repeated irradiations were carried out up to the 94th transfer generation. The dose was 1,000 r. each for the tumour-bearing rats of the 1st to 31st generation, and 2,000 r. each for those of the 32nd to 94th generation. The tumour cells of the pre-irradiated line were given an integral dose of 87,480 r.

X-ray irradiation of 2,000 r. appeared to be an almost lethal dose for the tumour cells of this type. Five to fifteen rats were used for each transplantation following irradiation. Those transplantations frequently failed. When successful, 7–18 days were required for tumour cell proliferation in the peritoneal cavity after transplantation. In the non-treated control line, on the other hand, 3–4 days were sufficient for the proliferation. Judging from the results[6] of a single cell inoculation of the Yoshida sarcoma into the peritoneal cavity, it is assumed that a few tumour cells remain to proliferate after X-irradiation of 2,000 r.

In order to compare the radiosensitivity of the tumour cells of the pre-irradiated line with that of the non-irradiated line, the cells of both sublines were transplanted subcutaneously in the right shoulder of rats. The tumour regression of both lines following local irradiation with 1,000 r. was studied. The factors of the irradiation were 185 kVp., 6 m.amp., half-value layer 0·76 mm. of copper, focus-tumour distance 13 cm., radiation field 4 cm. in diameter and dose rate 232 r./min. in air. The growth-rate of the tumours was studied by measuring size of tumours, regarding them as ellipsoids in shape. No difference was observed between two sublines. The grade of tumour-regression was higher for the pre-irradiated line than for the non-irradiated line following local irradiation with 1,000 r. This characteristic remained unaltered for 4 months without being exposed to X-rays. The mean values of the tumour size of both lines are shown in Fig. 1. Regression curves were statistically determined and significant difference was found between both sublines. The rate of cure of the subcutaneously transplanted tumours of the pre-irradiated line following local irradiation was significantly higher than that of the non-irradiated line.

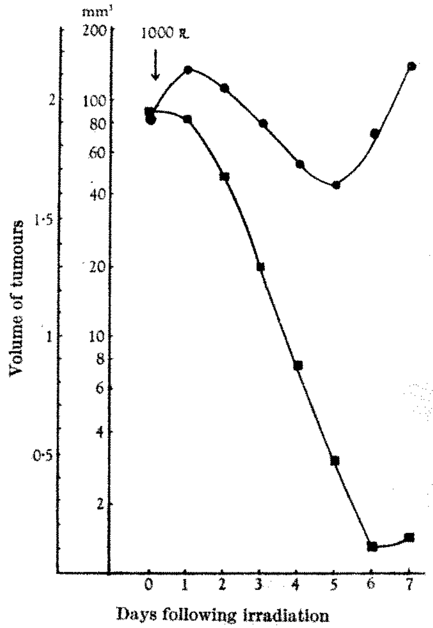

Fig. 1. Changes in volume of subcutaneously implanted tumours following local irradiation with 1,000 r. ●—●, Non-irradiated control line ($y = 1{\cdot}73514 - 0{\cdot}200679\,(x - \bar{x}) + 0{\cdot}00752355\,(x - \bar{x})^2 + 0{\cdot}0159303\,(x - \bar{x})^3$). ■—■, Pre-irradiated line ($y = 0{\cdot}860190 - 0{\cdot}459041\,(x - \bar{x}) + 0{\cdot}0155764\,(x - \bar{x})^2 + 0{\cdot}0178113\,(x - \bar{x})^3$)

From these results, it might be concluded that the pre-irradiated line is not more radioresistant, but more radiosensitive, than the control. It is quite conceivable that irradiation itself induces a mutation which increases the radiosensitivity of tumour cells. From the results of this experiment, it may be inferred that some of the homogeneous population of tumour cells may probably have undergone some mutational changes during the repeated heavy irradiations with X-rays and that these mutant cells proliferate as altered stem-cells. One of the biological characteristics of the pre-irradiated line is that tumour cells involved are more radiosensitive than those in the original line. The fact that selection of tumour cells was not observed may indicate that the individual tumour cell is almost equal in radiosensitivity in the Yoshida sarcoma.

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[1] Schubert, G., *Z. Krebsforschung*, **60**, 216 (1954).

[2] Dittrich, W., Höhne, G., and Schubert, G., in "Progress in Radiobiology", ed. by Mitchell, Holmes and Smith, 381 (Oliver and Boyd, 1955).

[3] Conger, A. D., and Luippold, H. J., *Cancer Res.*, **17**, 897 (1957).

[4] Brues, A. M., "Adv. Canc. Res.", **2**, 190 (1954).

[5] Windholz, F., *Radiol.*, **48**, 398 (1947).

[6] Sato, H., *Gann*, **43**, 1 (1952).

BIOLOGY

Transfer of Germ-Cells in *Xenopus laevis*

THE work of Bounoure[1] and Blackler[2] on three anuran species has shown that, in fixed and sectioned material, and after the application of suitable stains, the embryonic sites that contain primordial germ-cells may be recognized at all developmental stages from the fertilized egg to the tadpole. A technique for the transfer of the gonocytes between one embryo and another has been devised as a first step in a genetical analysis of abnormal embryos produced in this department by the transplantation of somatic nuclei into unfertilized eggs of *Xenopus*.

Three stages of development offer the most likely chances of successful transfer. The first of these is at the late blastula stage, in which the primordial germ-cells are localized in the superficial blastocœl floor. Second, at neurula stages, where the cells can be found in the ventral part of the posterior endoderm, and finally, at the stage 40 tadpole (Nieuwkoop and Faber's table[3]) in which the gonocytes are situated at the dorsal crest of the endoderm. Most successful transfers have so far been achieved at neurula stages.

The technique involves the attempted sterilization of the host embryo (though complete sterilization is not absolutely necessary for our purposes) by removal of its germ-cell site, and the replacement of this by the identical site from a donor embryo. Of course it is necessary to ensure that there is no possibility of confusing graft cells with any host germ-cells remaining after an unsuccessful or only partially successful sterilization. In this respect the use of the Oxford nuclear marker, discovered by Elsdale, Fischberg and Smith[4], has proved invaluable. In normal *Xenopus* embryos the nuclei of both somatic and germ-line cells possess potentially two nucleoli, though in some of the cells these nucleoli may be fused. Embryos bearing the mutation affecting nucleolar number possess but a single nucleolus in all nuclei. Thus when normal germ-cells are grafted into 1-nucleolate embryos and the host embryonic gonads later examined in microscopical sections, any cell present the nucleus of which bears two nucleoli must necessarily be of graft origin.

The method of transfer consists in decapsulating host and donor neurulæ in Niu and Twitty solution, excising the posterior endoderm of the host ventral to the archenteric cavity (together with its overlying ventral mesoderm and ectoderm) and replacing it by the same region taken from the donor embryo. The complete obliteration of the wound cuts by ectodermal overgrowth takes a variable time but operated embryos may eventually be transferred to 0·1 Niu and Twitty, in which solution they can be left for a couple of days. About 40 per cent of the operations are successful in that the graft heals in and the host embryo does not develop abnormally.

Results achieved so far have demonstrated clearly the presence in host gonads of grafted germ-cells, though the marker does not allow any conclusion to be reached in terms of 1-nucleolate germ-cells, which may be of host or graft origin. The latest stages to which these results apply at present are metamorphosed frogs in which, in females, the process of oocyte growth has begun.

Thus primordial germ-cells can be successfully grafted from one individual to another. However, it is not sufficient to prove only this point in establishing the technique, for the method is only of practical use if the grafted germ-cells are capable of giving rise to definitive and functional sex-cells. But if they do, then a central question inherent in the germ-cell problem in vertebrates will be settled, namely, as to whether or not primordial germ-cells give rise to definitive gametes. Such a result would be even more unassailable than the striking indications of germ-cell continuity arising from Mintz and Russell's[5] work on mouse embryos. Host animals are at present being reared to sexual maturity with the view of analysing the transfers by breeding, and it is hoped to publish a full account of the technique at a later date.

I wish to thank Dr. M. Fischberg for determining the nucleolarity of the embryos used. The work is being supported by a grant from the British Empire Cancer Campaign.

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[1] Bounoure, L., *Ann. Sci. Nat.*, 10e Ser., **17**, 67 (1934).

[2] Blackler, A. W., *J. Embryol. Exp. Morph.*, **6**, 491 (1958).

[3] Nieuwkoop, P. D., and Faber, J., "Normal Table of *Xenopus laevis*" (North-Holland Pub. Co., 1956).

[4] Elsdale, T. R., Fischberg, M., and Smith, S., *Exp. Cell. Res.*, **14**, 642 (1958).

[5] Mintz, B., and Russell, E. S., *J. Exp. Zool.*, **134**, 207 (1957).

Dermal Surface of Sheep-skin

In the dermal surface of sheep-skin a natural feature has been noticed, which is the origin of a defect in the appearance of some gloving leathers. During manufacture of leather the epidermis is removed, the exposed surface becoming the 'grain' of the leather, and small dark spots, scattered several mm. apart, occur on the grain surface of some leathers. A spot is illustrated in Fig. 1 (top), which shows the surface of a leather made from an Abyssinian sheep-skin. The rows of dark regions represent

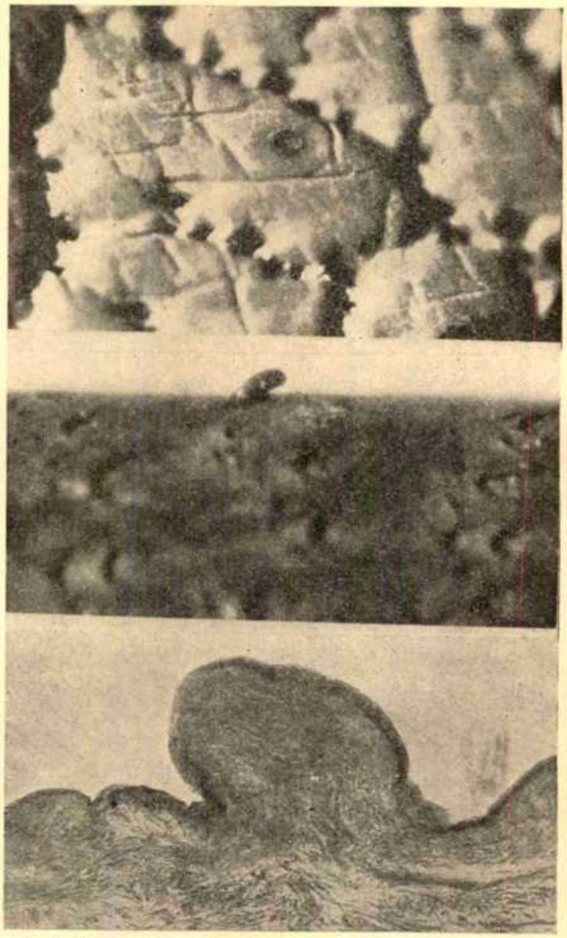

Fig. 1, Top, grain surface of gloving leather showing defective spot between two rows of hair follicle mouths (× *c*. 30); middle, 'tongue' rising from the grain surface of gloving leather (× *c*. 27); bottom, section of the base of a 'tongue' showing it to be a fibrous continuation of the dermis (× *c*. 188) the mouths of hair follicles and an offending spot lies between two such rows. Microscopical examination of cross-sections showed that the spot was not due to diseased or scar tissue, but that the surface connective tissue fibres had been damaged and were, in the leather, stuck together to give a glassy appearance to the spot. The examination of some other leathers of similar type, the origin of which was, however, not precisely known, gave a lead as to the cause of the spots. Fig. 1 (middle) shows a small tongue of tissue rising from the grain surface: the camera is looking along the grain surface so that the tongue is seen in profile. Fig. 1 (bottom) shows the base of a tongue in cross-section: it is fibrous and continuous with the general fibrous tissue of the dermis. The distribution of tongues was similar to that of the spots, and it seems reasonable to conclude that they rub off during processing, leaving the unsightly spots.

Similar spots or tongues have been observed, so far, in leathers from Abyssinian, Cape, Welsh Hill and Australian Merino sheep-skins.

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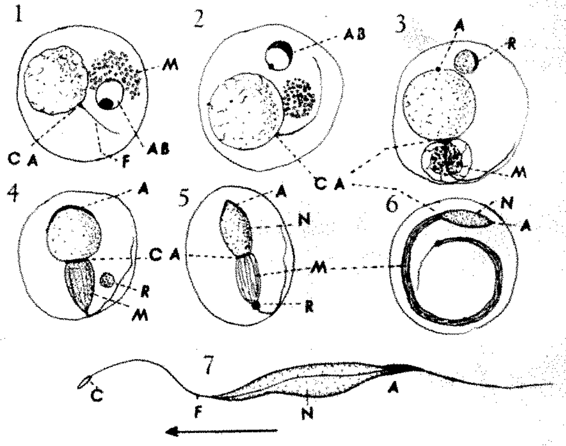
Position of the Proximal Centriole in Flagellate Spermatozoa

What must be one of the most remarkable spermatozoa of Insecta is that of *Lepisma*, as reported by Bowen[1], Nath and Bhatia[2], and Nath[3]. In Fig. 7, this spermatozoon is shown with a centriole (*C*) as its anterior end, connected by a flagellum (*F*), across the nucleus (*N*) to another body (*A*) said to be the acrosome, from which passes back the tail of the spermatozoon. With the acrosome in such a peculiar position, it is difficult to understand how this spermatozoon gets into the egg, always assuming that the acrosome of *Lepisma* takes the usual part in this process. It would seem also that the centriole lies in an unsheltered position, assuming again that this centriole is necessary for initiating the fertilization spindle of *Lepisma*.

We have recently studied the spermiogenesis of another thysanuran, *Petrobius maritimus*, and again are unable to support the interpretation given by Bowen and Nath. Nor do we believe that two insects so closely allied could have spermatozoa basically different in construction. In *Petrobius*, the acroblast (*AB*, Fig. 1) swells up in the early spermatid, producing the acrosome (*A*, Fig. 3), and afterwards flows down as the reject (*R*), which becomes attached for a time to the base of the mitochondrial nebenkern (*M*, Fig. 5). In Fig. 1 the flagellum (*F*) arises from a centriole-like body (*CA*) and curls around inside the cell; the body (*CA*) later forms a flat stainable plate (*CA*) between the nucleus and the mitochondrial middle piece (Fig. 4). This we believe to consist of an anterior centriole, and the centriole adjunct or neck body packing. The acrosome (*A*) lies in its normal position at the head end of the ripening spermatozoon (Fig. 6 *A*). No movement of the centriole forwards has been seen to occur in *Petrobius*. The question of the nature of the plate (*CA*) cannot be solved in such small cells by optical microscopy,



but the formation of the acrosome shown in Figs. 1–3 is quite normal, is within the powers of optical microscopy, and conforms to what has previously been seen in insect spermiogenesis. In some of our preparations, the nucleus between stage Figs. 4 and 5 appeared to have a lateral fold which we were unable to interpret, but which may be Bowen's supposed forward continuation of the flagellum (*F*, Fig. 7). Thus we consider that in Fig. 7, after Bowen and Nath, the anterior bead (*C*) is not the centriole, but is the acrosome (whole or part), whereas the body (*A*) is the adjunct plus the centriole, as has recently been shown to be the case in Orthoptera and Coleoptera by Gatenby and Tahmisian[4], who used the higher powers of the electron microscope.

In stages of spermiogenesis later than Fig. 6, the ripening sperm is seen to have double spiral threads passing down around the flagellum. The sperm is very small; but we could not find any reason to believe, as did Nath, that this spermatozoon is bereft of mitochondrial substance.

In his useful review on the cytology of spermatogenesis, Nath[3] discusses various "atypic flagellate sperms with centriole at the anterior tip". All such cases need re-investigation with the electron microscope, and until this has been carried out, we are inclined to believe that such "atypic flagellate spermatozoa" have no existence in the animal kingdom. Another statement which we find nowadays difficult to accept is that the acroblast directly metamorphoses into the acrosome, that is, "the direct method of acrosome formation" according to Nath and a few previous authors.

So far as we are able to understand, all recent work with the electron microscope supports the view that the acroblast is a pile of lamellæ (or more rarely a series of lamellated vesicles) which in some way concentrates the material included in the acrosome. It does not seem easy to believe that this pile of lamellæ changes into a globular electron translucent acrosome. On the contrary, the lamellæ always seem to be rejected as the Golgi remnant.

References to a centriole at the anterior end of the spermatozoon occur sporadically in the immense literature of the optical microscopy of animal spermiogenesis; but such recent electron microscopists as Burgos and Fawcett[6], Grassé, Carasso and Favard[7], Afzelius[8], Yasuzumi and Tanaka[9], Gatenby and Dalton[10], Gatenby and Tahmisian[4], who worked on mammals, molluscs, insects, oligochætes or seaurchins, all found the proximal centriole in its normal post-nuclear position in the neck of the spermatozoon.

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[1] Bowen, R. H., *J. Morph. Physiol.*, **39**, 351 (1924).
[2] Nath, V., and Bhatia, C. L., *Res. Bull. East Panjab Univ.*, **27**, 33 (1953).
[3] Nath, V., *Internat. Rev. Cytol.*, **5**, 395 (1956).
[4] Gatenby, J. Brontë, and Tahmisian, T. N., *La Cellule*, **59**, lx (1959).
[5] Gatenby, J. Brontë, and Mukerji, R. N., *Quart. J. Micro. Sci.*, **73** (1929).
[6] Burgos, M. H., and Fawcett, D. W., *J. Biophys. and Biochem. Cytol.*, **287** (1955).
[7] Grassé, P.-P., Carasso, N., and Favard, P., *Ann. Sci. Nat. Zool.*, **339** (1956).
[8] Afzelius, B., in Almquist and Wiksell, "Boktryck" (Uppsala, 1957).
[9] Yasuzumi and Tanaka, *J. Biophys. and Biochem. Cytol.*, **4**, 621 (1958).
[10] Gatenby, J. Brontë, and Dalton, A. J., *J. Biophys. and Biochem. Cytol.*, **6**, 45 (1959).

Mitochondria of Goldfish, *Carassius auratus*

The metabolism of mitochondria of poikilotherms acclimatized at different temperatures has not been studied, though such studies have been made on rats[1,2]. The mechanisms of adaptation to cold and warm temperatures in a poikilotherm may be different from those of a homoitherm due to the presence of a temperature-regulating centre in the latter. Hence, mitochondria from livers of goldfish, *Carassius auratus*, acclimatized at 10° and 30° C. were isolated to study their oxidative phosphorylation. The following account gives the method of isolation and the structure of mitochondria of the liver of unacclimatized goldfish.

The liver was placed in ice-cold 0·25 *M* sucrose solution immediately after its removal from the goldfish. All the following steps for the isolation of mitochondria were conducted at 0°–2° C. 2 gm. of the liver were homogenized in 10 c.c. of 0·25 *M* sucrose (buffered at *p*H 7·4 with *tris* buffer and containing 0·001 *M* versene) by fifteen up-and-down strokes of a Potter–Elvehjem homogenizer driven at 300 r.p.m. The homogenate was centrifuged at 700*g* for 10 min. in a refrigerated centrifuge to remove the cellular debris and nuclei. Then the supernatant was pipetted out and centrifuged at 14,000*g* for 10 min. to sediment the mitochondria.

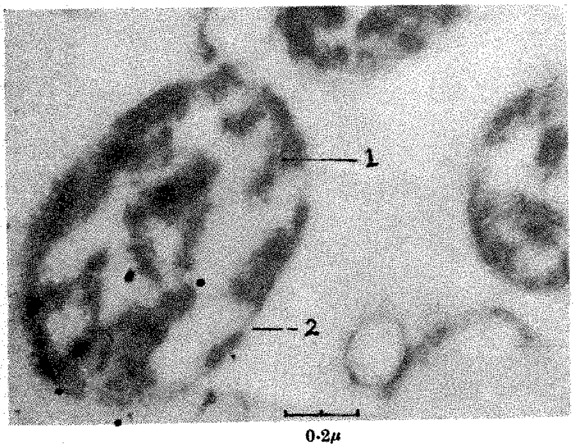

Fig. 1. Electronmicrograph of liver mitochondria of goldfish. 1, Cristæ; 2, double membrane. (× 40,000)

Electronmicrographs of the mitochondrial pellets showed that the mitochondria of the liver of goldfish obtained by the above method were intact (Fig. 1). They were oval in shape, 0·8–1·0μ in length and 0·5–0·6μ in breadth. They had a double membrane and cristæ as in rat liver mitochondria. These mitochondria stained well with janus green.

Oxidative phosphorylation of the liver mitochondria of goldfish acclimatized at 10° and 30° C. will be discussed elsewhere. I am grateful to Prof. C. L. Prosser, Department of Physiology, University of Illinois, for his guidance.

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[1] Panagos, S., Beyer, R. E., and Masoro, E. J., *Biochim. Biophys. Acta*, **29**, 204 (1958).
[2] Smith, R. E., and Fairhurst, A. S., *Proc. U.S. Nat. Acad. Sci.*, **44**, 705 (1958).

Spread of the Ascidian, *Styela mammiculata* Carlisle

Carlisle[1] described a new species of ascidian, *Styela mammiculata*, which was first taken in 1953, near Plymouth. If, as Carlisle suggested, this is an introduced species, it may be expected to spread. The discovery of numerous specimens on various submerged surfaces and in dredged material in Langston Harbour, Hampshire, during the summer of 1959, is therefore of interest. The species appears to be extending its range rapidly along the south coast of England, and increasing in numbers where already established. We expect it to spread to other sheltered and warm areas of the British and continental coast, either by natural means or by transport on the hulls of ships.

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[1] Carlisle, D. B., *J. Mar. Biol. Assoc. U.K.*, **33** (2), 329 (1954).

FOREST PRODUCTS

Effect of Moisture Changes on Creep in Wood

During studies on creep in wood under various types of loading over periods of up to several years, it became apparent that changes in the moisture content of the wood while under load markedly influenced both creep-rate and total creep.

To explore this further, small beams of ¾-in. square cross-section were cut from billets of three species, namely, *Eucalyptus regnans* (a hardwood prone to collapse of the cell wall during seasoning), *Eucalyptus pilularis* (a hardwood with very little tendency to collapse), and *Pinus radiata* (a softwood with negligible tendency to collapse). Three moisture conditions were studied, namely, green material maintained in the green state during test, initially green material allowed to dry while under load, and material air-dried to 12 per cent moisture content before loading and maintained at approximately that value during test.

The tests were conducted in a temperature-controlled atmosphere at 25° C. The beams were simply supported and subjected to four-point loading for a period of approximately two weeks. During this time the moisture content of the wood that was allowed to dry had reached a reasonably constant value. Unloaded beams of matched material were used as controls at each moisture condition. The maximum deflexions of the beams relative to the points of support, and the strains in the extreme fibres on the compression and tension faces near the centre of the span, were measured at intervals during test.

The results showed that the relative creep in wood allowed to dry while under load was at least twice that occurring in wood maintained at constant moisture content. Relative creep is defined as the ratio of creep deformation to initial deformation. Further, the relative creep was approximately the same in wood maintained at constant moisture content irrespective of whether it was green or dry. The strains in the extreme fibres were, for all three moisture conditions, much greater on the compression faces of the beams than on the tension faces, the maximum values of extreme fibre strains in both compression and tension being obtained for material drying under load. Similar results were obtained for each of the species tested.

Exploratory experiments have indicated that creep also increases markedly in wood subject to increasing moisture content during test, and that the effect of moisture changes on stress relaxation in beams held at constant deflexion show similar trends (P. U. A. Grossman, personal communication).

In the case of green wood drying while under load a marked difference in total creep and creep-rate has been observed in geometrically similar beams of different sizes. The average drying-rates for the sizes tested differed considerably and it would be expected that the restraints due to moisture gradients in the beams would have been different. Experiments are in progress to determine to what extent the rate of loss or gain of moisture affects the creep-rate and total creep.

In addition, tests have been commenced to determine the effect on creep of small changes in the moisture content of dry wood.

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PATHOLOGY

Urinary Selenium and Dental Caries

The suggested direct relationship between urinary selenium concentrations and the prevalence of dental caries in American children[1,2] is not substantiated by an investigation in New Zealand. Until recently, studies on the physiological effects of selenium have been concerned with its toxicity. Later evidence suggests that for some animals, and at certain levels, the element may be classified as essential[3]. Although selenium has not been demonstrated to be essential in man, its suspected role in electron transport may suggest a possible modification of the calcified structures. In New Zealand, the marked improvement in health, weight gain, and reduction of muscular dystrophy in sheep ('white muscle disease') by dosing with selenium[4], suggests there is a minimum level of available selenium in many areas of New Zealand. This evidence warranted an investigation of the supposed selenium–caries relationship.

Analytical data obtained in this investigation, and other work to be published elsewhere, indicate the low levels of available selenium.

The investigation was carried out in two rural areas of the South Island. Both areas are of different soil types, one being of yellow-grey and the other of yellow-brown earths. In neither district has selenium deficiency been reported in farm animals.

In South Otago, 39 school boys 5–14 years old were examined for dental conditions, and 31 of similar ages were examined in North Canterbury. The children were life-time residents of their districts.

All dental examinations were carried out by the one examiner (P. B. C.), and were supplemented by bitewing radiographs. 24-hr. samples of urine were collected under parental supervision, from the children concerned. Samples were collected in polythene bottles containing toluene as a preservative. Every practicable care was taken to ensure the reliability of the samples.

Selenium analysis was carried out by a modification of the colorimetric diamino-benzedine method of Luke[5]. Recoveries were of the order of 94 per cent. Readings were made on Beckman model *DU* spectrophotometer.

Selenium was present in all urine samples. In Table 1 the mean values of urinary selenium-levels for both groups are lower than those in the four areas investigated by Hadjimarkos[1,2], and are believed to be indicative of low selenium availability. The difference of the means for the districts is significant at the 99 per cent level of confidence.

Table 1. Distribution of Urinary Selenium-Levels

Urinary selenium (p.p.m.)	Number of male subjects 5–14 years old in :	
	South Otago	North Canterbury
0·00–0·09	2	0
0·10–0·19	18	3
0·20–0·29	13	11
0·30–0·39	5	15
0·40–0·49	1	2
Totals :	39	31
Means :	0·021 p.p.m.	0·030 p.p.m.
Standard deviation :	± 0·009	± 0·008

It has been shown that the rate of urinary excretion is relatively constant, and is quite independent of urinary volume[6]. As the concentration of selenium is a function of the volume excreted, the volume being subject to climate, fluid intake, and other factors, it is felt that a more reliable basis of comparison may be given by the total urinary selenium excreted in 24 hr.

Table 2 gives the amount of selenium excreted by subjects in the two districts in terms of µgm./24 hr. The mean excretions in each district are significantly different at the 98–99 per cent level of confidence.

The percentage of available tooth surfaces decayed and filled was 22·6 at South Otago and 18·8 at North Canterbury. The difference of 3·8 has a standard error of 9·8. Therefore, the difference is not significant.

There was no correlation between the proportion of available tooth surfaces decayed and filled and the total urinary selenium excreted in 24 hr. (Correlation coefficients were plus 0·019 for South Otago, and plus 0·06 for North Canterbury. Standard errors of the correlation coefficients were 0·16 and 0·16, respectively).

Table 2. SELENIUM EXCRETED IN μGM./24 HR.

Selenium (μgm./24 hr.)	Number of male subjects 5–14 years old in: South Otago	North Canterbury
0–9	5	1
9·1–18	21	10
18·1–27	9	11
27·1–36	3	6
36·1–45	0	1
45·1–54	0	0
54·1–63	1	2
Totals	39	31
Means	17·0 μgm.	23·7 μgm.
Standard deviation	± 9·8	± 12·2

In this investigation the significant difference in selenium excretion between groups was not associated with any significant difference in caries prevalence.

This investigation was carried out as a project of the dental research unit of the Medical Research Council of New Zealand. The assistance and advice of Captain F. L. Losee (D.C.), U.S.N., is gratefully acknowledged.

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[1] Hadjimarkos, D. M., and others, *J. Pediatrics*, **40**, 451 (1952).

[2] Hadjimarkos, D. M., and Bornhorst, C. W., *J. Pediatrics*, **52**, 274 (1958).

[3] Schwarz, K., and Doltz, C. M., *J. Amer. Chem. Soc.*, **79**, 3292 (1957).

[4] Hartley, W. J., *N.Z. J. Agric.*, **99**, 259 (1959).

[5] Luke, C. L., *Anal. Chem.*, **31**, 572 (1959).

[6] Sterner, J. H., and Lidfeldt, V., *J. Pharm. Therap.*, **73**, 205 (1941).

Cementing Substances in Metastasizing and Non-Metastasizing Transplantable Tumours in Mice

VERY little is known about whether, or how far, the condition of intercellular cementing substances as present in the tissue of a growth may be responsible for local invasion and for metastasis[1,2]. The situation is all the more complex as neoplastic cells may even themselves produce polysaccharides[3], which are considered to be the fundamental chemical constituents of the intercellular cement. The condition of the cementing substances in 13 transplantable tumours in mice have been studied in this laboratory. Results are summarized in Table 1.

Primary tumours were fixed in fluids containing alcohol, formalin and picric and acetic acids, preferably at low temperature (Duboscq–Brasil and Gendre). Particular emphasis was placed on selecting non-necrotic portions for microscopical study. Several staining and histochemical procedures for identifying mucopolysaccharides were used, the most useful being the Hale[4,5] and the periodic acid–Schiff reactions. Phloxine and hæmalum were also occasionally used for cytoplasmic and nuclear counterstaining. The latter removes, however, the Hale-positive cement of epitheliomas and should not be employed in neoplasms of this type.

(1) *The ground substance in eight transplantable sarcomas.* The greatest quantity of the Hale-positive material was present in the ground substance of non-metastasizing tumours (see Table 1, 1–4). There was in these tumours also a great amount of alcian-blue-positive material (not given in Table 1). On the other hand, the Hale-positive material was absent in highly and moderately metastasizing tumours (Table 1, 7 and 8); the quantity was intermediate in slightly metastasizing tumours (Table 1, 5 and 6). In other words, metastatic capacity increases when Hale-positive material diminishes. By contrast, the periodic acid–Schiff-positive material was absent in most of non-metastasizing tumours (Table 1, 1, 2 and 3) and present in most of metastasizing tumours (Table 1, 5, 6 and 8). But there were definite exceptions from this rule (Table 1, 4 and 7). These exceptions are most probably due to the technique, which does not prevent a certain degree of extraction of the more soluble components of periodic acid–Schiff.

Digestion by hyaluronidase and metachromatic staining tend decidedly to favour the assumption that Hale-positive material is chemically related to mucopolysaccharides, at least in certain non-metastasizing sarcomata.

(2) *The intercellular cementing substances in five epitheliomas.* The antimetastatic influence of Hale-positive material was again fully evident in epitheliomas (Table 1, 9–13). On the other hand, metastatic capacity was the more pronounced the greater the quantity of periodic acid–Schiff (compare Table 1, 9 with 10–13). A moderate amount of a similar

Table 1

No.	Tumour*	Strain of mouse	Metastatic capacity	Amount of Hale-positive material in the: Ground substance‡	Intercellular cement‡	Cytoplasm	Amount of periodic acid – Schiff in the: Ground substance	Intercellular cement	Cytoplasm
	Sarcomas								
1	*Sa*†	*DBA*/1	none	+ + + +		+	−		trace
2	*Sb*†	*DBA*/1	none	+ + + +		+	−		—
3	*GL*46 (early transfer)	*DBA*/1	none	+ + +		trace	−		trace
4	*D*3†	*C3H*	none	+ + +		trace	+ +		trace
5	*GL*46 (later transfer)	*DBA*/1	slight	+ +		trace	+		trace
6	*S*180	*C57BL*/10*d*	slight	+		—	trace		trace
7	*D*1†	*C3H*	moderate	−		trace	−		trace
8	*T*241	*C57BL*	high	−		trace	+		trace
	Epitheliomas								
9	*Pch* (solid)† ●	*CA*58	slight		+	+ +		−	−
10	*K*7†	*C3H*	moderate		+	+		+	−
11	*K*9†	*C3H*	moderate		+	+		+	−
12	*Pch* (ascitic)†	*CA*58	moderate		trace	trace		+	+ +
13	*T*150	*DBA*/1 or *Lp*	high		−	−		+	trace
12′	*Pch* (ascitic) (*ip*)†	*CA*58	slight		+	+ +		trace	+

● All epitheliomas, excepting the bladder carcinoma 13, are mammary adenocarcinomas.

* All were grafted subcutaneously. In addition, *Pch* (ascitic) was inoculated intraperitoneally (see 12′).

† Isografts; 5, 6 and 13 are homografts which took in 100 per cent of cases.

‡ Quantities of ground substance in sarcomas and of intercellular cement in epitheliomas are not comparable; comparison is admissible only within the same type of tumours, that is, within the group of sarcomas, or within the group of epitheliomas.

substance could also be seen in the stroma of the more invasive epitheliomas (not mentioned in Table 1), a phenomenon already described by Gersh and Catchpole[2].

(3) *Intracytoplasmic Hale- and periodic acid–Schiff-positive material.* Hale-positive material was in general present in the cells of slightly and non-metastasizing tumours (Table 1, 1, 2, 9 and 12′); it was absent or less abundant in the cells of other metastatic tumours (exception Table 1, 10 and 11). The intracytoplasmic periodic acid–Schiff was observed in diffuse form in all cells, and as granules in some. The granules were apparently absent in tumours 2, 9, 10 and 11.

Cytoplasmic Hale-positive material frequently occupied a concavity of the nucleus (Golgi zone). This is probably the site of synthesis of Hale-positive material, especially in the ascitic form of *Pch* (Table 1, 12′). The concentration of mitochondria around this region is in favour of this assumption, as likewise various morphological signs of metabolic activity of the nucleus: digitations of its membrane and a greater concentration of the chromatin adjacent to the Golgi zone.

In several tumours (Table 1, 1, 2, 9, 10, 11 and 12′) cytoplasmic Hale-positive material seemed to be continuous with that present in the intercellular spaces. A similar continuity of periodic acid–Schiff material was observed only occasionally and irregularly.

Certain observations suggest that the cementing substances are synthesized and secreted rather than absorbed by the tumour cells. This assumption is supported by the extension of the cytoplasmic Hale-positive material from the nuclear indentation to the cell wall, and finally to intercellular spaces, as observed in our tumour 12′ growing intraperitoneally. This may possibly explain why tumours differ as to local invasiveness and metastatic behaviour, both depending seemingly on the quantity and nature of the cementing substances originating in the tumour cells themselves.

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[1] Ambrose, E. J., *The Advancement of Science*, No. 57, 473 (June 1958).
[2] Gersh, I., and Catchpole, H. R., *Amer. J. Anat.*, **85**, 457 (1949).
[3] Meyer, K., "Harvey Lectures", **61**, 88 (1957).
[4] Müller, G., *Acta Histochem.*, **2**, 68 (1955).
[5] Graumann, W., and Clauss, W., *Acta Histochem.*, **6**, 1 (1958).

Relation of Leydig Cells in the Human Testicle to the Tubules and Testicular Function

GROUPS of Leydig cells have been found in the spermatic cords in 23 of 30 consecutive necropsies. The groups of Leydig cells most commonly occur in the 3 cm. of cord immediately above the upper pole of the testes, but they may be found up to 10 cm. above the globus major, with considerable variation in distribution, both in numbers present and numbers of levels. The cells are constantly distributed in the loose areolar tissue around nerve sheaths and usually infiltrate the nerve bundle, disorganizing the characteristic wavy pattern of nerve fibres. Such cells show all the morphological features characteristic of Leydig cells, crystalloids of Reinke and hyaline bodies occasionally being evident. Infiltration of the nerves by Leydig cells has been described, but not explained, by various authors[1-4]. Some thirty years ago this peculiar association occasioned discussion as to the nature of the cells[5,6]. The relation to nerves can be accounted for either (*a*) by migration along nerve fibres at the embryonic period when nerve fibres grow into testicular parenchyma, or (*b*) by traction of cells from the formed parenchyma during descent of the testicle. The first explanation can be discounted by the similarity of the trabecular pattern of capillaries in normal nerves and the trabecular pattern shown by the infiltrative cells before their continued proliferation disrupts the organized appearance of the nerve.

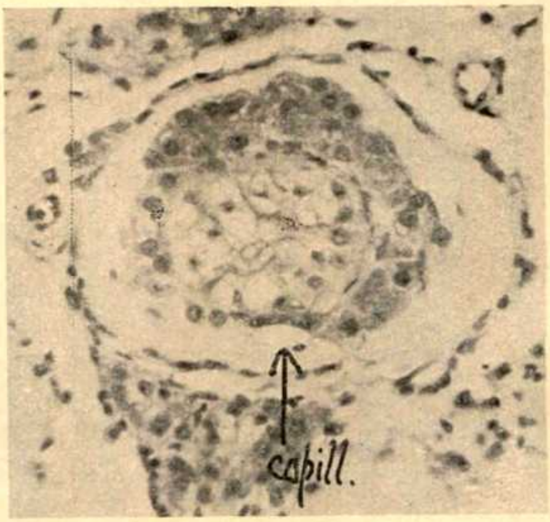

Fig. 1. Hyaline thickening of tubule with residual germinal epithelium and intact inner lamella surrounded by Leydig cells, and with empty capillary spaces at 6 o'clock. (Hæmatoxylin and eosin. × 250)

A similar infiltrative proliferation is seen in hyaline change of the tubular substantia propria where histological detail is clearer, and where active changes take place at the inner lamella, to which are applied thin-walled capillaries. In ordinary paraffin preparations such capillaries are one cell thick and similar to those inside nerve bundles. From the earliest phases of hyaline thickening of the tubule wall, varying numbers of Leydig cells have been seen within the substantia often continuous with groups outside the tubule. In advanced thickening, Leydig cells are typically applied to the inner lamella (Fig. 1). Leydig cells are generally regarded as being an endocrine epithelium and intimate application to thin-walled capillaries is to be anticipated. This association in sites that are abnormal by conventional anatomical standards may be of general biological significance.

In the testes, the intertubular tissue is of scant cellularity, with most of the capillaries present, either in Leydig cell groups or closely applied to the tubule wall. Thus, there are few available capillaries of optimum size along which Leydig cells may proliferate, and the immediate explanation of this infiltrative activity probably is that the capillaries of the invaded structures are the only ones available. This raises the questions (*a*) why, in Leydig cell proliferation, adequate numbers of new capillaries are not formed, and (*b*) what factors limit growth along the existing vascular tree.

In (*a*) a recent preliminary report[7] on the possible reduction in the number of small blood vessels in the seminal vessels of the castrate rat is in part suggestive that the local action of Leydig cell secretion may effect capillary proliferation. However, the process of infiltration of the hyaline substantia is usually diffusely distributed. In (*b*) if it is assumed that endocrine epithelium must have a thin-walled capillary blood supply, it follows that limitation of proliferation along the capillary bed, away from the tubular capillaries, will be effected by increasing thickness of the vessel wall, be it arteriole or venule. The point of importance in these observations is that the same capillary supplies Leydig cells and tubule.

Because of the extreme tortuosity of the tubules and the close application of many of the Leydig cells to the tubular wall, it is difficult to demonstrate clearly on which side of the tubular capillary bed the Leydig cells are situated. In the spermatic cord the Leydig cells are situated close to the arterioles and the same relations probably exist within the testicular parenchyma.

On the basis of these findings, I advance the hypothesis that Leydig cells are situated between the arterioles and the testicular tubules and have the same capillary blood supply as the tubules. This hypothesis may usefully be applied to several aspects of testicular function, notably the gonadotrophic effect of steroid hormones and their varying effect at different dosage-levels, the irregular segmental distribution of atrophic changes in mature testes, pubertal development and abnormal segmental development as in cryptorchidism, as well as possible mechanisms in male infertility and its therapy.

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[1] Nelson, A. A., *Amer. J. Path.*, **14**, 831 (1938).
[2] Sternberg, W. H., *Amer. J. Path.*, **25**, 493 (1949).
[3] Okkels, H., and Sand, K., *J. Endocrinol.*, **2**, 38 (1940).
[4] Warren, S., and Olshausen, K. W., *Amer. J. Path.*, **19**, 307 (1943).
[5] Brannen, D., *Amer. J. Path.*, **3**, 343 (1927).
[6] Berger, L., *C.R. Acad. Sci., Paris*, **175**, 907 (1922).
[7] Berry, R., *et al.*, *Angiology*, **10**, 372 (1959).

HISTOLOGY

Histochemical Demonstration of Succinic Dehydrogenase Activity in Osteoclasts

A REVIEW of the literature has revealed but few studies on enzyme characteristics of osteoclasts. None of these, however, related to dehydrogenase activity.

In the present work frozen sections of rat and hamster knee joints and jaws, 8–10μ thick, were prepared according to the Adamstone–Taylor cold knife technique and mounted on glass microscope slides. In addition, pieces of calvarium from new-born hamsters were also employed. Slides and specimens were then incubated for 60–90 min. in the succinic dehydrogenase substrate solution as described by Pearson[1]. This contained nitroneotetrazolium. Representative sections were also incubated in a substrate solution containing nitro blue tetrazolium[2] as described by Nachlas *et al.* With both substrates an intense selective staining reaction was observed in osteoclasts (Fig. 1). Sections incubated in tetrazole without the co-factor (succinate) were unstained, as were heat-treated sections. Sodium malonate was inhibitory. The staining of osteoclasts associated with the remodelling resorption of the jaws was particularly striking. Osteoblasts by contrast showed little if any reaction.

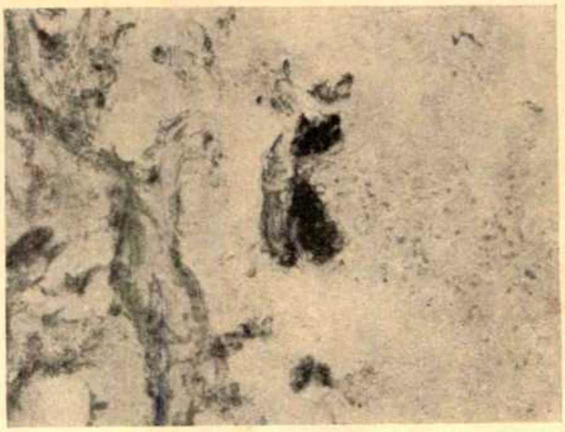
Fig. 1. Succinic dehydrogenase activity of osteoclasts (knee joint from 2-day-old hamster). (× *c*. 214)

Osteoclasts associated with both intramembranous and intracartilagenous bone resorption showed a similar reaction. The cytoplasm was evenly stained, and it was difficult to assess any mitochondrial reaction.

Osteoclasts have also been observed to exhibit high acid phosphatase[3] and aminopeptidase activity[4]. The present study would further tend to confirm the hypothesis that these cells exhibit high metabolic activity. Localization of succinic dehydrogenase to osteoclasts may be considered as presumptive evidence that the Krebs cycle is involved in the resorptive process.

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[1] Pearson, B., *J. Histochem. and Cytochem.*, **6**, 112 (1958).
[2] Nachlas, M. M., Tsou, K. C., De Souza, E., Cheng, C. S., and Seligman, A. M., *J. Histochem. and Cytochem.*, **5**, 420 (1957).
[3] Burstone, M. S., *J. Histochem. and Cytochem.*, **7**, 39 (1959).
[4] Burstone, M. S., Amer. Assoc. Adv. Sci. Monograph (in the press).

Histochemical Localization of Oxidase Activity in the Mitochondria of the Heart in Some Lower Vertebrates

WITH the aid of some new methods[1-3] the histochemical localization of oxidase activity was investigated in the mitochondria of the heart in some lower vertebrates, namely, the viviparous cyprinodonts, *Lebistes reticulatus*, *Xiphophorus helleri* and *Xiphophorus maculatus*, the carp, *Cyprinus carpio*, the frog, *Rana temporaria*, the toad, *Bufo calamita* and the lizard, *Lacerta agilis*.

To permit a satisfactory comparison with the results of Burstone[3] obtained with the mitochondria of the human heart, we applied the corresponding histological technique. Immediately after death by decapitation, 8-μ thick sections of frozen heart muscle were prepared according to the cold-knife method of Adamstone–Taylor, after which they were mounted on chemically clean glass microscope slides. The incubation, which was carried out at 24° C. and lasted ½ hr., was in a substrate solution containing 10 mgm. of variamine blue *B* base or *p*-methoxy-N-phenyl-*p*-phenylenediamine (Carbic Color and Chemical Co., New York) and 10 mgm. of *p*-aminodiphenylamine or N-phenyl-*p*-phenylenediamine (British Drug Houses, Poole, England). At first the substrates were placed in a 50·0-ml. Erlenmeyer flask and dissolved in 0·5 ml. of ethanol reagent. After adding 35·0 ml. of distilled water and 15·0 ml. of 0·2 *M tris* buffer *p*H 7·4 the solution was shaken and filtered through folded filter paper into a Coplin jar. Incubating solutions containing 0·001 *M* sodium sulphide as well as 0·001 *M* potassium cyanide were also used. In addition, the effect of adding per Coplin jar 20·0 mgm. of cytochrome *c* (Sigma Chemical Co., St. Louis, Mo.) to the substrate solution was studied. Some sections were pretreated with physiological saline for half an hour prior to the incubation. Afterwards the sections were incubated in substrate solutions with and without cytochrome *c*. After incubation the slides were stored for 1 hr. in a 10 per cent solution of cobaltous acetate in 10 per cent formalin containing 5·0 ml. 0·2 *M p*H 5·2 acetate buffer. Then the slides were washed for about 5 min. in running water and mounted in polyvinyl pyrrolidone[4] or glycerol-gelatin.

The distribution of the dye with routine incubations appeared to correspond to that found with the classical mitochondrial stains such as the Regaud stain. Just as in the human heart[3] the localization corresponded exactly to those observed for heart sarcomes or mitochondria by Cleland and Slater[5] when using the mitochondrial method of Altmann. Also in our cases the distribution of the staining corresponded very well with the sarcoma pattern as observed under the electron microscope[6]. Some areas showed a perinuclear staining corresponding to the localization of the perinuclear sarcomes.

When the reaction was completely inhibited by addition of sulphide and cyanide, as effected by pre-incubation in physiological saline, it was completely restored in the saline-treated sections by addition of cytochrome *c* to the substrate solution. Moreover, the staining intensity could be augmented to some degree by adding cytochrome *c* to the routine incubation solutions. Consequently, the cytochrome oxidase is believed to be associated with the mitochondria. Just as in the human heart[3], on the strength of the localization of the staining reaction to the mitochondria, the results of the sulphide and cyanide inhibition and the cytochrome *c* tests, one can conclude that cytochrome oxidase occurs in the heart of the lower vertebrates investigated.

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[1] Burstone, M. S., *J. Histochem. and Cytochem.*, 7, 112 (1959).
[2] Burstone, M. S., *J. Histochem. and Cytochem.* (in the press).
[3] Burstone, M. S., *Nature*, 184, 476 (1959)
[4] Burstone, M. S., *Amer. J. Clin. Path.*, 28, 429 (1957).
[5] Cleland, K. W., and Slater, E. C., *Quart. J. Micro. Sci.*, 94, 329 (1953).
[6] Kisch, B., and Bardet, J. M., "The Electron Microscopic Histology of the Heart" (Brooklyn Medical Press, New York, 1951).

ENTOMOLOGY

Nocturnal Detection of Tsetse Flies in Nigeria with Ultra-Violet Light

Jewell[1,2], in Tanganyika, showed that tsetse flies marked with fluorescent paint can be located at night by means of ultra-violet light resting on leaves. A portable petrol generator was his current supply (personal communication). Because such equipment is cumbersome, other workers[3] have used a hand torch to search for flies painted with reflective glass beads mixed with glue.

The ultra-violet method has been used at the West African Institute for Trypanosomiasis Research to study the nocturnal resting sites of *Glossina palpalis* R.-D. and *G. submorsitans* Newst. in Northern Nigeria. A portable, fairly compact, battery-powered current supply is used. In the case of *G. palpalis*, the information so obtained will form the basis of an experiment on the selective spraying of residual insecticide during the dry season.

The apparatus was supplied by Electric and Musical Industries, Limited, Hayes, Middlesex. A 12-V. car battery feeds d.c. current to a 'Valradio 12/150*B*' convertor which produces a.c. current at 250 volts. After passing through special control gear and the necessary chokes and condensers, this voltage powers two 80-watt ultra-violet bulbs, each in a parabolic reflector on 100 ft. of flex. The entire machine, in a wooden case, fits into a car boot. If no road exists to the study area, the equipment can be broken into two 50-lb. loads, one of which is the battery. These are easily head-carried by labourers. The pigment, a non-radioactive, luminescent powder, was supplied by Barber Medico-electronic Laboratories, Ltd., Weston-super-Mare, Somerset. It adheres well to the thorax of the fly if mixed with thin gum arabic office glue to which has been added a trace of detergent. In the laboratory, painted flies survive as well as unpainted controls. Four colours (blue, yellow, white, orange) are used, permitting sex, time of marking and hunger stage to be identified if desired.

The quantity of light varies with individual bulbs, the best of which concentrate a central beam of ultra-violet sufficient for detecting flies at a distance of 25–30 ft. Sufficient light in the visual range is produced to enable the operator to move about without a torch. The light is attractive to flies disturbed by the operator's movements, and some will fly into the reflector. These can be shaken out and if the lamp is directed away from them, will come to rest nearby. Undisturbed flies, however, do not leave their perches to come to the light, nor does closer inspection with a hand torch disturb them. Some confusion is caused by fireflies (Lampyridae), but these can be identified by their regular flashing. Certain arachnids and fungi fluoresce under ultra-violet, but never as brightly as the painted flies.

Many trials with the riverine species, *G. palpalis*, have revealed that about half the flies rest during the night within 12 in. of the ground, on leaves or small twigs, but never on tree trunks or branches. There appears to be no difference between the sexes in resting height preference at night. Flies released in the morning are rarely found on the night of the same day, having dispersed up- and down-stream in search of food. The best method is to catch the flies in the afternoon, then mark and drop them into a large cage with a removable top. At dusk the cage top is removed, and those flies that fly out are considered uninjured by handling and marking. 10–25 per cent of marked *G. palpalis* come to rest within the 125-ft. searching radius of the machine, the great majority of them alighting very close to the point of release.

Approximately 50 recoveries of marked specimens of the open woodland species, *G. submorsitans*, in a forest-island breeding site, revealed the flies resting on twigs, leaves and small creepers at heights ranging from 1 to 15 ft., with the majority above 7 ft. It would appear, then, that this species' daytime resting preference for tree trunks, at least when gorged[4], should be considered when planning control by residual insecticides.

Both species' habit of resting on leaves and twigs at night might be a means of avoiding predators such as ants and jumping spiders, the vibration of the predator's approach serving to alert the fly, whereas a branch or tree trunk daytime habitat would permit maximum visibility in the fly's search for a host.

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[1] Jewell, G. R., *Nature*, 178, 750 (1956).
[2] Jewell, G. R., *Nature*, 181, 1354 (1958).
[3] Rennison, B. D., *et al.*, *Nature*, 181, 1354 (1958).
[4] Nash, T. A. M., *Bull. Ent. Res.*, 43, 33 (1952).

Sex-limited DDT-Resistance in House-flies

A NEW mechanism of inheritance of DDT-resistance has been investigated in a selected strain of house-flies (*Musca domestica* L.) wherein resistance is manifest only in males. Genetic tests have shown that the resistance is transmitted by males but not by females. It is therefore determined either directly by a *Y*-chromosome factor or, like sex in *Drosophila*, by the balance between the unpaired parts of *X* and *Y* and the rest of the chromosome set.

Evidence for the existence of this patrilineal inheritance dates back to 1954 when an attempt was first made to derive a homogeneous susceptible strain from the Canberra laboratory colony[1] which, although bred since 1939 in isolation from insecticides, had been found to contain a DDT-resistance factor in a small proportion of both males and females. Selection for early maturation (giving strain *E*) eliminated resistant females by the sixth generation, but the strain continued to produce some resistant males which, during a further 68 generations, increased in proportion to about 75 per cent of males produced.

At this stage a complete separation of resistant and susceptible males was effected in a single selection by breeding, on one hand, from males which survived a topically applied dose of 32·0 μgm. DDT/gm. body-weight, and, on the other hand, from males which when tested later were killed by a dose of 11·2 μgm. DDT/gm. The two strains so obtained were designated *EY* (early, with resistance involving *Y*) and *ES* (early, susceptible).

Males and females of *ES* and females of *EY* constitute three similar and apparently homogeneous populations in respect to their tolerances to DDT, which lie between 2·0 and 11·2 μgm./gm. *EY* males are consistently about eight times as resistant, their tolerances ranging from 16·0 to 128·0 μgm./gm.

Strain *EY* has bred true without any applied pressure since the initial selection. All its males are specifically DDT-resistant and all its females non-resistant. It contrasts markedly with previously described strains of houseflies, for in these resistance to DDT has been found to be autosomally determined and not sex-linked or sex-limited[2-4]. A fuller account of this work will be published elsewhere.

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[1] Kerr, R. W., Venables, D. G., Roulston, W. J., and Schnitzerling, H. J., *Nature*, 180, 1132 (1957).
[2] Milani, R., *Riv. Paras.*, 17, 233 (1956); 18, 43 (1957).
[3] Crow, J. F., *Ann. Rev. Ent.*, 2, 227 (1957).
[4] Brown, A. W. A., World Health Organization Monograph Series No. 38 (1958).

BACTERIOLOGY

Intermediary Metabolism of Nitrobenzoic Acids by Bacteria

KE *et al.*[1] have recently reported that the aerobic degradation of *o*-nitrobenzoic acid by a *Flavobacterium* involves the *o*-nitroso and *o*-hydroxylamino compounds. The corresponding amino-derivative, anthranilic acid, was not oxidized, their paper implying on the basis solely of manometric data that it was not a potential metabolite of *o*-nitrobenzoic acid although they found that cells grown on anthranilic acid could oxidize both it and salicylic acid. We wish to direct attention to the fact that although the aminobenzoates may not always be directly concerned in aerobic degradation of nitrobenzoic acids, they are nevertheless important intermediary metabolites.

We had previously recorded identical responses to those reported by Ke *et al.*, with *p*-nitrobenzoic acid and its derivatives, using cells of *Nocardia erythropolis* grown in the presence of *p*-nitrobenzoic acid, and with *o*-nitrobenzoic acid using cells of *N. opaca*, grown in a medium containing *o*-nitrobenzoic acid[2]. Although the aminobenzoic acid in both cases was not appreciably oxidized, it nevertheless accumulated in culture media. Work with cell-free extracts has shown that in addition to oxidation of the substrate, *N. erythropolis*, at least, carries out an appreciable, simultaneous reduction of the *para*-isomer to *p*-aminobenzoic acid by a pathway involving *p*-nitrosobenzoic and *p*-hydroxylaminobenzoic acids[3].

In contrast to Durham's earlier work[4,5] and the recent report of Ke *et al* [1], no further oxidation of the aminobenzoates through the corresponding hydroxybenzoates has been found with our species of *Nocardia*. Cells of *N. opaca* grown on anthranilic acid failed to oxidize salicylate and growth of *N. erythropolis* was not maintained on *p*-aminobenzoic acid[6]. The use of desiccator-dried preparations of *N. opaca* showed that permeability factors were not responsible for absence of activity in that particular case.

The scheme of Ke *et al.*[1] whereby *o*-hydroxylaminobenzoic acid is metabolized to cellular material without passing through anthranilic acid is interesting in view of some results we obtained with *N. erythropolis*, which oxidizes *p*-nitrobenzoic acid through *p*-hydroxybenzoic acid to protocatechuic acid, also without the mediation of the amino-compound.

Table 1. PRODUCTION OF PROTOCATECHUIC ACID FROM *p*-NITROBENZOIC ACID AND ITS REDUCTION PRODUCTS BY *N. erythropolis*

Substrate	Protocatechuic acid produced		
	Exp. 1*	Exp. 2*	Exp. 3*
p-Nitrobenzoic acid	++++	++++	+++
p-Nitrosobenzoic acid	±	±	–
p-Hydroxylaminobenzoic acid	+++	+++	+
p-Aminobenzoic acid	–	–	–
None	–	–	–

–, ±, +, ++, etc., Intensity of protocatechuic acid spots estimated visually.

* Identical sheets were marked in the areas corresponding to protocatechuic acid, the areas cut out, eluted and a full spectrum read (210–320 mμ).

Experiment 1 was made with 24-hr. grown cells; experiment 2 with 48-hr. grown cells and experiment 3 with cells grown for 7 days, on *p*-nitrobenzoate.

Washed cell suspensions of *N. erythropolis* (grown on *p*-nitrobenzoate medium) in *tris* buffer with low aeration rates (see Cartwright and Cain[2] for full experimental details of this and all other methods used) produced appreciable amounts of protocatechuic acid from both *p*-nitro and *p*-hydroxylaminobenzoic acids, but much smaller amounts from *p*-nitrosobenzoic acid and none from the amino-compound. It was detected chromatographically in three solvent systems, identified spectrophotometrically (Table 1) and also isolated in pure form from a *p*-nitrobenzoate incubation mixture. The *p*-hydroxylaminobenzoic acid sample used was recrystallized several times from water prior to use and was shown to be quite free of nitro-compound by paper chromatography.

The results would suggest a pathway from *p*-hydroxylaminobenzoic acid to protocatechuic acid and terminal oxidation products lacking an intermediate amino-stage and support the results of Ke *et al.* with the *ortho*-isomer. Although *p*-nitro, *p*-nitroso and *p*-hydroxylaminobenzoic acids were oxidized at similar rates by cells grown on *p*-nitrobenzoic acid, the effects of inhibitors (particularly 1 m*M* nitrite) and other findings[2] still lead us to maintain that, with *Nocardia*, nitrite is the immediate nitrogenous product and that rapid reduction to ammonia is achieved by nitrite reductase activity. This reflects, as Ke *et al.*[1] have pointed out, the great diversity of pathways by which different micro-organisms may degrade the same substrate, but we must advocate the necessity for carrying out concomitant chemical studies with adaptation results of this kind. Our own results have shown that although *p*-aminobenzoic acid is not a direct intermediate in the primary aerobic, energy-producing pathway of *p*-nitrobenzoic acid degradation, it is eventually metabolized both in *p*-nitrobenzoate cultures and after incubation with washed cells of *N. erythropolis* grown on *p*-nitrobenzoate[3]. We have since identified further aminophenyl compounds produced in cultures from *p*-nitrobenzoate and from *o*-nitrobenzoate together with the corresponding hydroxynitrobenzoic acids (Table 2) and detected at least six other unidentified phenolic compounds from the former, none of which is oxidized by the organisms. Recent work (which will be reported in full elsewhere) on anthranilic acid metabolism by *N. opaca* confirms the multiplicity of compounds to which this aminobenzoate may be converted in cultures. This, we think, serves to emphasize the imprudence of eliminating a compound as a major metabolite merely from adaptation results based on manometric studies. If not involved in aerobic pathways, it may well be important in others.

Table 2. SOME AMINOPHENYL AND RELATED METABOLITES PRODUCED BY *Nocardia* SPP. FROM NITROBENZOIC ACIDS

Substrate	Species	Metabolites identified	First appears in cultures (days)
p-Nitrobenzoic acid	*Nocardia erythropolis*	*p*-aminobenzoic acid	1
		3-hydroxy-4-nitrobenzoic acid	3
		3-hydroxy-4-aminobenzoic acid	3
o-Nitrobenzoic acid	*N. opaca*	3-hydroxy-2-nitrobenzoic acid	3
		3-hydroxyanthranilic acid	3
		anthranilic acid	½
		o-aminophenol	3–4

These metabolites were found both in cultures of the two species and in incubation mixtures of washed cells of the respective species in 0·05 *M tris* or phosphate buffer, *p*H 7·2, with *p*- or *o*-nitrobenzoic acid at 0·01 *M*. Incubation temperature in both cases was 30°.

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[1] Ke, Y-H., Lynn, L. G., and Durham, N. N., *J. Bacteriol.*, 77, 593 (1959).
[2] Cartwright, N. J., and Cain, R. B., *Biochem. J.*, 71, 248 (1959).
[3] Cartwright, N. J., and Cain, R. B., *Biochem. J.*, 73, 305 (1959).
[4] Durham, N. N., *J. Bacteriol.*, 72, 333 (1956).
[5] Durham, N. N., *Canad. J. Microbiol.*, 4, 141 (1958).
[6] Cain, R. B., *J. Gen. Microbiol.*, 19, 1 (1958).

Intrastrain Transduction in *Proteus mirabilis*

A NUMBER of bacteriophage-mediated transduction systems have been described[1]. Intrastrain transduction was originally described for *Salmonella typhimurium* by Zinder and Lederberg[2] and recently Loutit has described a similar situation in *Pseudomonas aeruginosa*[3].

In an attempt to discover transducing systems in the genus *Proteus* for use in genetic transfer between *vulgaris* and *mirabilis* species, lysogeny in this genus was investigated[4]. Two of the temperate phages isolated[5] have been successfully employed in intrastrain transductions. The first was present in the supernatant of 10-day-old cultures of *Proteus mirabilis* strain 34[6] and formed turbid plaques on *Proteus mirabilis* strain 13; it is named phage 34/13. Phage 12/57 was obtained after ultra-violet irradiation[5] of *Proteus mirabilis* strain 12; it forms turbid plaques on *Proteus mirabilis* strain 57. The two transducing systems are analogous and as results are very similar only the one employing phage 12/57 will be described.

Streptomycin-resistant mutants (57/*S*) of the wild type 57 were obtained by plating about 10^{11} cells on a MacConkey-type agar[7] containing 1,000 μgm. streptomycin sulphate/ml. Colonies which arose were picked off and purified by re-streaking on a streptomycin medium.

Phage 12/57 does not produce clearing of broth cultures of 57 or 57/*S*, and in order to obtain high-titre lysates, recourse was taken to the modification of the confluent plaque method of Hershey *et al.*[8] described by Adams[9]. Successful experiments produced lysates of 57 or 57/*S* with plaque titres of 10^{10}/ml. as estimated by the agar layer method[10]. Lysates were centrifuged to clarity and rendered bacteriologically sterile by addition of chloroform.

Transduction experiments were conducted in a 37° C. water-bath and consisted of mixing 2 ml. of a 57/*S* lysate (about 2×10^{10} plaque-forming particles) with a deposit of about 6×10^{8} wild type 57 cells obtained by centrifuging 1 ml. of an aerated overnight culture. The experiments were duplicated using a lysate of the wild type 57 of identical titre. No visible clearing occurs, and 15 min. after mixing, 0·1 ml. volumes were spread on a number of Difco *SS* agar plates. At 30-min. intervals 0·1 ml. volumes were spread on MacConkey agar containing 1,000 μgm. streptomycin sulphate/ml. After overnight incubation the *SS* plates were replicated[11] on to MacConkey agar containing 1,000 μgm. streptomycin sulphate/ml. An organism control (6×10^{8} wild-type 57 in 2 ml. broth) and a lysate sterility control (2 ml. of the lysate) were similarly treated.

Replica plates of the *SS* agar in experiments using lysate from 57/*S* usually yielded about 100 colonies per plate, while those in experiments using the lysate from wild 57 were always sterile. Replica plates from organism controls were usually barren but occasionally showed 1 or 2 colonies per experiment. Lysate controls invariably confirmed their bacteriological sterility.

In experiments using lysates from 57/*S* the half-hourly direct platings on MacConkey agar containing 1,000 μgm. streptomycin sulphate/ml. first yielded colonies 2 hr. after mixing organisms and lysate. The corresponding plates in experiments using lysates of wild 57, and in the organism and lysate controls, never showed growth.

The transducing activity of 57/*S* lysates is not impaired by treatment with deoxyribonuclease (40 μgm. per ml. for 2 hr. at 37° C.) but is totally inhibited by phage 12/57 rabbit antiserum, which reduces the plaque titre from 10^{10} to 10^{4}/ml. The addition of a similar volume of normal rabbit serum to the lysate has no such effect.

Results outlined place this experiment in the class of bacteriophage-transmitted intrastrain genetic exchange.

The streptomycin resistance marker has been the only one employed to date. The high mutation-rate of wild 57 and wild 13 to sodium azide resistance precludes the use of this property as a marker. No biochemical or nutritional mutants have yet been recovered.

The transduction frequency per phage particle is about 10^{-7}, and is comparable with rates encountered in common transduction systems[12]. There appears to be a delay in phenotypic expression of transduced streptomycin resistance. This delay has also been encountered with the transduction of streptomycin resistance in the *Salmonella typhimurium* system[13]. As in the latter system no abortive transductions have been detected.

Study of the transduced organisms has proved that the streptomycin resistance is a stable characteristic. These organisms do not differ biochemically from the wild type. None of the colonies tested to date adsorbs phage present in either 57 or 57/*S* lysates. Mutants of wild 57 resistant to phage present in lysates of 57 are not resistant to streptomycin. The mechanism by which the streptomycin-resistant transductants of 57 become resistant to phage is being investigated.

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[1] Stocker, B. A. D., in "Recent Progress in Microbiology", ed. by Tunevall, G., 31 (Blackwell Scientific Publications, Oxford, 1959).
[2] Zinder, N. D., and Lederberg, J., *J. Bact.*, **64**, 679 (1952).
[3] Loutit, J. S., *J. Gen. Microbiol.*, **18**, 315 (1958).
[4] Coetzee, J. N., and Sacks, T. G., *Nature*, **184**, 1340 (1959).
[5] Coetzee, J. N., and Sacks, T. G., *S. Afr. J. Lab. Clin. Med.* (in the press).
[6] Coetzee, J. N., *S. Afr. J. Lab. Clin. Med.*, **4**, 147 (1958).
[7] Coetzee, J. N., *S. Afr. J. Lab. Clin. Med.*, **5**, 16 (1959).
[8] Hershey, A. D., Kalmanson, G., and Bromfenbrenner, J., *J. Immunol.*, **46**, 267 (1943).
[9] Adams, M. H., in "Methods in Medical Research", ed. by Comroe, J. H., **2**, 12 (The Year Book Publishers, Chicago, 1956).
[10] Adams, M. H., in "Methods in Medical Research", ed. by Comroe, J. H., **2**, 7 (The Year Book Publishers, Chicago, 1956).
[11] Lederberg, J., and Lederberg, E. M., *J. Bact.*, **63**, 399 (1952).
[12] Bertani, G., in "Advances in Virus Research", ed. by Smith, K. M., and Laufer, M. A., **5**, 182 (Academic Press, New York, 1958).
[13] Watanabe, T., and Watanabe, M., *J. Gen. Microbiol.*, **21**, 16 (1959).

Characteristics of *Thiobacillus thioparus*

DURING investigations of the bacterial decomposition of thiosulphate and thiocyanate in effluents from gasworks, it became necessary to identify the organisms responsible. These were *Thiobacilli*, and one in particular, organism *G*, appeared identical with *T. thioparus* as described in Bergey[1]. To confirm this, its behaviour was compared with various *Thiobacilli* from the National Collection of Industrial Bacteria. As a result of this comparison it was discovered that the type species, *T. thioparus* (NCIB 8370), did not correspond with Bergey's classification on two important points: it grew on nutrient agar, and it grew anaerobically on thiosulphate containing potassium nitrate. To check this a further specimen of the organism was obtained, and although this was again a pure culture it showed precisely the same characteristics.

After discussion with a member of the staff of the National Chemical Laboratory, it was concluded that due to difficulties associated with the culture originally given to the Collection, NCIB 8370 was probably not *T. thioparus*.

The matter is of some importance, as strain NCIB 8370 was used by De Kruyff *et al.*[2] to show that *T. thioparus* and *T. thiocyanoxidans* are identical; as it seems not to be *T. thioparus* their conclusions would appear to be invalid.

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[1] Bergey's "Manual of Determinative Bacteriology", 7th ed. (Baillière, Tindall and Cox, 1957).
[2] De Kruyff, C. D., van der Walt, J. P., and Schwartz, H. M., "Antonie van Leeuwenhoek", **23**, 305 (1957).

FORTHCOMING EVENTS

(*Meeting marked with an asterisk * is open to the public*)

Monday, March 21

Society of Chemical Industry, Pesticides Group (at 14 Belgrave Square, London, S.W.1), at 5.30 p.m.—Dr. E. K. Woodford and Mr. L. Kasasian: "Some New Herbicides and Developments in Chemical Weed Control 1958–60".

Manchester Literary and Philosophical Society (in the Reynolds Hall, Manchester College of Science and Technology, Sackville Street, Manchester), at 5.45 p.m.—Mr. C. E. Young: "The Growing Importance of Metallo-Organic Compounds" (Presidential Address).

Institution of Mechanical Engineers, Industrial Administration and Engineering Production Group (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Informal Discussion on "The Computer in Production".

Institution of Electrical Engineers, London Graduate and Student Section (at Savoy Place, London, W.C 2), at 6.30 p.m.—Dr. F. A. Benson: "Developments in Lighting".

Royal Geographical Society (at 1 Kensington Gore, London, S.W.7), at 8.30 p.m.—Mr. Martin W. Holdgate: "Islands of Western Patagonia".

Tuesday, March 22

British Museum (Natural History) (in the Lecture Hall, Cromwell Road, London, S.W.7), at 4.30 p.m.—Prof. Charles G. Sibley (Cornell University): "Genes, Proteins and Evolution—The Phylogeny of Birds as indicated by Molecular Structure".*

Institute of Physics, Electronics Group (at 47 Belgrave Square, London, S W.1), at 5.30 p.m.—Annual General Meeting, followed by Mr. P. O. Hawkins: "Neutron Sources".

Institution of Chemical Engineers (at the Geological Society, Burlington House, Piccadilly, London, W.1), at 5.30 p.m.—Mr. R. L. Brown and Mr. J. C. Richardson. "Profile of Flow of Granules Through Apertures".

Tuesday, March 22—Thursday, March 24

Royal Institution of Naval Architects (in the Weir Lecture Hall, 10 Upper Belgrave Street, London, S.W.1), at 10.15 a.m. daily —Spring Meeting.

Wednesday, March 23

Institute of Fuel (at the Institution of Civil Engineers, Great George Street, London, S.W.1), at 5 30 p.m.—Mr. H. R. Gregory: "A New Method of Briquetting Coal without a Binder".

Institution of Electrical Engineers, Electronics and Communications Section (at Savoy Place, London, W.C.2), at 5.30 p.m. —Mr. G. Millington: "The Challenge of the Propagation Medium to the Radio Engineer".

Royal Photographic Society, Scientific and Technical Group (joint meeting with the Colour Group of the Physical Society, in the Lecture Theatre of the Science Museum, South Kensington, London, S.W.7), at 5.30 p.m.—Mr. M. H. Wilson: "Two-colour Projection Phenomena".

Society of Chemical Industry, Food Group (at the Courtauld Hall, Queen Elizabeth College, Campden Hill Road, London, W.8), at 6 p.m.—Scientific Papers (Eighth Young Members' Meeting).

British Psychological Society, Medical and Social Psychology Sections (at the Royal Society of Medicine, 1 Wimpole Street, London, W.1), at 8.30 p.m.—Dr. R. Money-Kyrle and Dr. Marie Jahoda: "Prejudice".

Thursday, March 24

Royal Society (at Burlington House, Piccadilly, London, W 1), at 4.15 p.m.—Meeting for the Election of Fellows. Mr. A. G. Swallow and Miss Mary R. Truter: "The Stereochemistry of β-Diketo Complexes with Trimethylplatinum IV. I: The Crystal Structure of Trimethyl 4 : 6-Dioxononylplatinum"; Mr. A. C. Hazell and Miss Mary R. Truter: "The Stereochemistry of β-Diketo Complexes with Trimethylplatinum IV. II: The Crystal Structure of Ethyl (Trimethylplatini-)acetoacetate".

Institution of Mechanical Engineers, Applied Mechanics Group (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m. —Discussion on "A Replica Technique for Measuring Static Strains".

Royal Aeronautical Society (at the Institution of Mechanical Engineers, 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m. —Dr. R. R. Jamison and Mr. R. J. Lane: "Engines for Supersonic Air Liners".

Thursday, March 24—Friday, March 25

National Association for Mental Health (at Church House, Westminster, London, S.W.1)—Annual Conference—"Mental Health at Home and Abroad".

Friday, March 25

Royal Aeronautical Society (at 4 Hamilton Place, London, W.1), at 7 p.m.—Mr. D. Perkins: "Aerodynamics of Man Powered Flight, with special reference to Inflatable Wings".

Royal Institution (at 21 Albemarle Street, London, W.1), at 9 p.m.—Prof. Ronald King: "Tyndall, The Teacher".

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Cowburn Research Fellow, to undertake research on certain aspects of immunology in relation to cancer—The Secretary, King's College Hospital Medical School, Denmark Hill, London, S.E.5 (April 2).

Assistant Experimental Officer (with a science degree preferably including bacteriology and chemistry, and some experience in bacteriological work) in the Bacteriology Department—The Secretary, Hannah Dairy Research Institute, Kirkhill, Ayr (April 4).

Assistant Lecturer in Physiology or Pharmacology—The Registrar, The University, Nottingham (April 4).

Senior Lecturer in Applied Thermodynamics; a Senior Lecturer in Fluid Mechanics; a Senior Lecturer in Mechanics of Machines; a Senior Lecturer in Strengths of Materials; a Senior Lecturer in Production Technology; and a Lecturer in Mechanical Engineering—The Registrar, Royal Technical College, Salford 5, Lancs (April 4).

Assistant (preferably with special interests in the biochemistry or micro-organisms, or in physical biochemistry) in Biochemistry —The Secretary of University Court, The University, Glasgow (April 8).

Assistant (with a degree in agriculture, horticulture or economics) to the Horticultural Liaison Officer in the Department of Agricultural Economics—The Registrar, The University, Nottingham (April 9).

Assistant Lecturer (preferably with a knowledge of some branch of comparative physiology, especially neurophysiology) in the Department of Zoology—The Registrar, University College of Wales, Aberystwyth (April 9).

Lecturer (with a good honours degree in electrical engineering or the equivalent, and ability to participate effectively in research work in this field) in the Department of Electrical Engineering, with special reference to the transmission and distribution of electric power—Prof. A. Tustin, Department of Electrical Engineering, Imperial College of Science and Technology, London, S W.7 (April 9).

Senior Lecturer in Virology, to be responsible for the organization of virology and for teaching in the Department of Pathology —The Secretary, Medical College of St. Bartholomew's Hospital, West Smithfield, London, E.C.1 (April 9).

Lecturer in Agricultural Chemistry—The Secretary of University Court, The University, Glasgow (April 11).

Lecturer in Astronomy in St. Salvator's College—Joint Clerk to the University Court, College Gate, St. Andrews, Fife (April 16).

University Lecturers (2) (one with special reference to mycology and plant pathology and the other to ecology) in Botany—Dr. C. B. Goodhart, Museum of Zoology, The University, Downing Street, Cambridge (April 18).

Cancer Research Fellow (with training in cytology), for duties mainly in connexion with the study of human chromosomes in patients with leukæmia and associated diseases—The Registrar, The University, Liverpool, quoting Ref CV/N (April 19).

Chair of Zoology in the University College of Ghana—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (April 20).

Senior Lecturer or Lecturer in Anatomy; and a Senior Lecturer or Lecturer in Physiology at the University of Khartoum—The Registrar, University of Khartoum, c/o Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (April 20).

Lecturer/Senior Lecturer (with either travel or field experience in Greece and Italy or the Near East) in Classical or Near Eastern Archæology, at the University of Sydney, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, April 22).

Assistant Lecturer (with special qualifications in physical chemistry) in Chemistry—The Secretary, The Royal College of Science and Technology, George Street, Glasgow, C.1 (April 23)

Assistant Lecturer or Lecturer (preferably with a special interest in some group of fossils common to the lower Palæozoic rocks) in Palæontology—The Registrar, University College of Wales, Aberystwyth (April 25)

Imperial Chemical Industries Research Fellows in Biochemistry, Chemistry, Chemotherapy, Engineering, Metallurgy, Pharmacology or Physics—The Academic Registrar, University of London, Senate House, London, W.C.1 (April 30).

Lecturer (graduate in science) in Microbiology—The Secretary, The Queen's University, Belfast (April 30).

Lecturer in Inorganic Chemistry—The Registrar, The University, Manchester 13 (April 30).

Research Assistant (with an honours degree in mathematics or a related subject, and preferably experience in the use of digital computers) in the Computing Laboratory—The Secretary of University Court, The University, Glasgow (April 30).

Senior Lecturer in Physics; and a Lecturer in Physics at the University of Natal, Pietermaritzburg, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa and London, April 30).

Turner and Newell Research Fellow in Engineering, Inorganic Chemistry or Physics—The Academic Registrar, University of London, Senate House, London, W.C.1 (April 30).

University Students and Fellows, for experimental or theoretical research in nuclear and elementary particle physics—The Professor of Natural Philosophy, The University, Glasgow (April 30).

Chair of Chemistry in the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, May 30).

Biochemist (interested in biological assay work) for Endocrinology Laboratory, working in connexion with cancer research—The House Governor, Royal Marsden Hospital, Fulham Road, London, S.W.3.

Biochemist (preferably with previous hospital experience) in the Pathological Laboratory—The Secretary, Highlands General Hospital, Winchmore Hill, London, N.21.

CHEMIST or AGRICULTURAL CHEMIST (with an honours degree in science or equivalent qualification, and preferably postgraduate research experience to Ph.D. level, and a knowledge of agriculture), to undertake research on the quality of protein in animal feeding stuffs for non-ruminants—The Secretary, The Rowett Research Institute, Bucksburn, Aberdeen.

CYTOLOGIST (Scientific or Senior Scientific Officer grade) AT THE WELSH PLANT BREEDING STATION—The Registrar, University College of Wales, Aberystwyth.

DARWIN RESEARCH FELLOW (with suitable qualifications in science or medicine), for research in the field of human population genetics with special reference to the range of human variability—The Eugenics Society, 69 Eccleston Square, London, S.W.1.

EDITOR (chemist, with editorial experience) OF *Chemistry and Industry*—The General Secretary, Society of Chemical Industry, 14 Belgrave Square, London, S.W.1, marking envelope "Personal".

LECTURER (with good academic qualifications and either experience of, or special interest in, nuclear physics) IN PHYSICS, at Bournemouth Municipal College of Technology and Commerce, to help with the development of courses in nuclear and reactor physics and teach physics to Grad.Inst.P., H.N.C. applied physics and B.Sc. (General) levels—Chief Education Officer, Town Hall, Bournemouth.

MASTER TO TEACH CHEMISTRY throughout the School—The Headmaster, Malvern College, Malvern.

MICROBIOLOGIST (with research experience), for work on radiation-induced mutations in microbial populations—The Administrator, Radiobiological Research Unit, Medical Research Council, Harwell, Berks.

PHYSICISTS (with doctorate degree, and preferably a fair knowledge of French)—Le Directeur, Departement de Physique, Université de Montréal, Canada.

RESEARCH ASSISTANT (graduate), to join a group working on problems of cancer research in the border-line fields of biology and chemistry—The Administrative Officer, Mount Vernon Hospital, Northwood, Middlesex.

RESEARCH POST IN THE RADIOASTRONOMY GROUP, to participate in an expanding programme of research in upper atmosphere physics, including investigations of meteors, auroræ and solar-terrestrial relations—Dr. T. R. Kaiser, Department of Physics, The University, Sheffield 10.

TECHNICIAN or JUNIOR TECHNICIAN (interested in biochemistry), for biochemical and biological assay work in the ENDOCRINOLOGY LABORATORY—The House Governor, Royal Marsden Hospital, Fulham Road, London, S.W.3.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

The Registrar General's Statistical Review of England and Wales for the year 1957. Part 3: Commentary. Pp. xiv+299. (London: H.M. Stationery Office, 1959.) 16*s*. 6*d*. net. [3012

Time Measurement in Dynamic Geometry. By S. Ryzewski. Pp. 16. (Southampton: S. Ryzewski, 15 Landguard Road, 1959.) [3012

Africa: Social Change and Mental Health. (Report of a Panel Discussion conducted in Conference Room No. 1, United Nations, New York, March 23, 1959, by the World Federation for Mental Health). Pp. 40. (London: World Federation for Mental Health, 1959. Distributed by H. K. Lewis and Co., Ltd.) 3*s*. 6*d*. net. [3012

Privy Council. Medical Research Council Memorandum No. 37: Sewage Contamination of Bathing Beaches in England and Wales. Pp. iv+24. (London: H.M. Stationery Office, 1959.) 2*s*. 6*d*. net. [3012

National Museum of Wales. Fifty-second Annual Report, 1958–59. Pp. 79+4 plates. (Cardiff: National Museum of Wales, 1959.) [61

Review of Coal Tar Technology, January–June, 1959. (Vol. 11, Part 1.) Pp. ii+88. (Gomersal, Leeds: Coal Tar Research Association, 1959.) 10*s*. [61

Institute of Personnel Management. Occasional Papers, No. 15: Working Wives—a Survey of Facts and Opinions concerning the Gainful Employment of Married Women in Britain. Carried out in Co-operation with Mass Observation, Ltd. by Viola Klein. Pp. 63. (London: Institute of Personnel Management, 1960.) 7*s*. 6*d*. [61

Iodine Tests and Reagents in Chemical Analysis. Pp. i+52. (London: Chilean Iodine Educational Bureau, 1959.) [61

Bulletin of the British Museum (Natural History). Historical Series. Vol. 2, No. 2: Darwin's Notebooks on Transmutation of Species. Part 1: First Notebook (July 1837–February 1838). Edited with an Introduction and Notes by Sir Gavin de Beer. Pp. 23–73. (London: British Museum (Natural History), 1960.) 20*s*. [61

Annual Report of the Challenger Society, 3, No. XI. Pp. 38. (Wormley, Godalming: Challenger Society, c/o National Institute of Oceanography, R. I. Currie, Hon. Secretary, 1959.) [61

Annual Report of the National Oceanographic Council, 1 April 1958–31 March 1959. Pp. v+41. (Cambridge: At the University Press, 1959.) 5*s*. net. [61

British Broadcasting Corporation. The Public and the Programmes. an Audience Research Report on Listeners and Viewers, the time they devote to listening and viewing, the Services they patronize, their Selectiveness and their Tastes. Pp. 71. (London: British Broadcasting Corporation, 1959.) [61

Building Research Station Digest No. 120: Cranes for Building. Pp. 6. (London: H.M. Stationery Office, 1959.) 4*d*. [61

Ministry of Agriculture, Fisheries and Food. Bulletin No. 77: Tomatoes—Cultivation, Diseases and Pests. Fifth edition. Pp. v+74+8 plates. (London: H.M. Stationery Office, 1959.) 5*s*. 6*d*. net. [61

Scientific Proceedings of the Royal Dublin Society. Series A, Vol. 1, No. 1: Radiocarbon Dating System Using Scintillation Techniques. By C. F. G. Delaney and I. R. McAuley. Pp. 1–20. (Dublin. Royal Dublin Society, 1959.) 4*s*. [61

Economic Proceedings of the Royal Dublin Society. Vol. 4, No. 7: Observations for Eleven Years on a Liming Trial. By P. Bergin. Pp. 119–136. 4*s*. Vol. 4, No. 8: Chemical and Growth Gradients of *Laminaria cloustoni* Edm. (*L. Hyperborea* Fosl.) By W. A. P. Black, W. D. Richardson and F. T. Walker. Pp. 137–149. 3*s*. (Dublin. Royal Dublin Society, 1959.) [61

Ciba (A.R.L.), Limited. Technical Notes, No. 204 (December 1959) 'Araldite' in Slip Ring Assemblies. Pp. 8. (Duxford: Ciba (A.R.L.), Ltd., 1959.) [61

United Kingdom Atomic Energy Authority: Research Group Report (AERE—R3159): Heat Transfer to Two Phase Gas-Liquid Systems. Part 1: Steam-Water Mixtures in the Liquid-Dispersed Region in an Annulus. By J. A. R. Bennett, J. G. Collier, H. R. C. Pratt and J. D. Thornton. Pp. iv+46+13 figures. Report (AERE—R3195): Pressure-Drop Data for the Vertical Flow of Air-Water Mixtures in the Climbing-Film and Liquid-Dispersed Regions. By J. A. R. Bennett and J. D. Thornton. Pp. ii+22+6 figures. (Harwell Chemical Engineering Division, Atomic Energy Research Establishment, 1959.) [61

Horniman Museum and Library. A Handbook to the Collections Illustrating a Survey of the Animal Kingdom. Third edition. Pp. 92+4 plates. (London: London County Council, 1959.) 2*s*. 6*d*. [61

Northern Ireland Development Council. Freight Services to and from Northern Ireland. Second edition. Pp. i+42+6 plates. (London: Northern Ireland Development Council, 1960.) [61

Department of Scientific and Industrial Research. Summary of Progress of the Geological Survey of Great Britain and the Museum of Practical Geology for the year 1958, with the Report of the Geological Survey Board. Pp. iii+77. (London: H.M. Stationery Office, 1959.) 5*s*. net. [61

The National Union of Teachers. University Entrance—The Basic Facts. Pp. 48. (London: National Union of Teachers, 1960.) 3*s*. 6*d*. [61

British Medical Bulletin, Vol. 16, No. 1 (January 1960): Antibiotics in Medicine. Pp. 1–88+xvii. (London: The British Council, 1960.) 20*s*. [61

International Commission for Uniform Methods of Sugar Analysis. Report of the Proceedings of the Twelfth Session held at the National Bureau of Standards and at the Shoreham Hotel, Washington, D.C., U.S.A., from June 2nd to June 6th, 1958. Pp. vii+130. (Keston, Kent: International Commission for Uniform Methods of Sugar Analysis, Tate and Lyle Research Laboratories; New York: Sugar Research Foundation, Inc., 52 Wall Street, 1959.) 25*s*.; 3.50 dollars. [61

Colour Breeding of Syrian Hamsters. By Roy Robinson. Pp. 16. (Ealing, London: Robin Bibliographies, St. Stephens Road Nursery, 1960.) 3*s*. 6*d*. [121

Other Countries

State of Singapore. Fisheries Bulletin No. 8: Fisheries Survey Report No. 2—The Fishing Gear of the State of Singapore. By T. W. Burdon. Pp. vi+166+10 plates. (Singapore: Government Publications Bureau, 1959.) 2.50 Malayan dollars. [3012

Institut pour la Recherche Scientifique en Afrique Centrale. Onzième Rapport Annuel, 1958. Pp. 375. (Bruxelles: Institut pour la Recherche Scientifique en Afrique Centrale, 1959.) [3012

Colony of Mauritius. Meteorological Observations and Climatological Summaries, 1958. Part 7: July. Pp. 14. Part 8: August. Pp. 14. Part 9: September. Pp. 14. Part 10: October. Pp. 14. Part 11: November. Pp. 14. Part 12: December. Pp. 25. (Port Louis: Government Printer, 1959.) [3012

Canada: Department of Mines and Technical Surveys. Geological Survey of Canada. Bulletin No. 51. Surficial Geology of Southern District of Keewatin and the Keewatin Ice Divide, Northwest Territories. By Hulbert A. Lee. Pp. vi+42 (13 plates). 50 cents. Memoir No. 298: Coalfields, West Half Cumberland County, Nova Scotia. Pp. vi+89. 75 cents. (Ottawa: Queen's Printer, 1959.) [61

La Science en Pologne Populaire, 1944–1959. Par Tadaesz Kotarbinski. (Rapport présenté à l'Assemblée Générale de l'Académie Polonaise des Sciences le 26 juin 1959.) Pp. 45. (Varsovie: Editions Scientifiques Polonaises, 1959.) [61

United States Department of Commerce: Coast and Geodetic Survey. Technical Bulletin No. 7: Pantograph Adjustment. By G. C. Tewinkel. Pp. ii+3 5 cents. Technical Bulletin No. 10: Vertical Adjustment of Instrument Aerotriangulation by Computational Methods. By William D. Harris. Pp. ii+8. 10 cents. (Washington, D.C.: Government Printing Office, 1959.) [61

Union of South Africa. Department of Commerce and Industries. Division of Fisheries Investigational Report No. 34: The Cape Fur Seal (*Arctocephalus pusillus*)—Distribution Abundance and Feeding Habits off the South Western Coast of the Cape Province. By R. W. Rand. Pp. 75. (Reprint from *Commerce and Industry*). (Pretoria: Government Printer, 1959.) [61

University of Khartoum: Natural History Museum. Annual Report for period 1st July, 1956 to 30th June, 1958. Pp. 19. (Khartoum: University of Khartoum: Sudan Natural History Museum, 1959.) [61

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IN THE SERVICE OF YOUTH

THE second[*] of two recently published reports concerned with the way in which the State should assist in the service of youth has met with much more sympathetic public response than the first[†]. The Committee was set up by the Minister of Education just over a year ago, under the chairmanship of the Countess of Albemarle, and has completed its inquiry with exemplary promptness. Invited to "review the contribution which the Youth Service of England and Wales can make in assisting young people to play their part in the life of the community, in the light of changing social and industrial conditions and of current trends in other branches of the education service ; and to advise according to what priorities best value can be obtained for the money spent", the Committee quickly realized that it had been convened none too soon and that speedy action would be required if the Youth Service were to flourish. It is not surprising to learn from the Report that the Committee was always aware that it was working against time and that the recommendations made should be regarded as in the nature of a blood transfusion. Side by side with the Committee's proposals for immediate adoption come proposals which would ensure sustained and continued nourishment if the blood transfusion succeeds in restoring the health of a sickly and dispirited child.

The main factor contributing to the sense of urgency which governed the Committee's inquiry is, of course, the 'bulge'. For every five young people between the ages of fifteen and twenty in Britain to-day there will be, in 1964, six young people. Although the increase will not be spread evenly across the country as a whole, all areas and each kind of area will show some increase ; in some new towns the increase may be as much as five-fold. Between 1964 and 1970 the total number will decline gradually, but will still remain substantially higher than that of to-day. From 1970 there is again the probability of a gradual increase. The inadequate facilities which are available in the Youth Service to-day are quantitatively insufficient to meet demands which are already upon us. Measures taken to meet the impact of the 'bulge' in the schools and technical colleges and the universities have not been matched with similar provision to prepare the Youth Service for the needs of the increased population. The Committee quickly recognized that unless action is taken at once the 'bulge' generations will leave school only to find a Youth Service inadequate to cope even with its earlier responsibilities.

The effects of the 'bulge' will be reinforced by the cessation of National Service. During the past few years military commitments have kept about 200,000 young men between the ages of eighteen and twenty out of civilian life. Its gradual abandonment during the next two or three years will leave this number of young men available for civilian employment and leisure pursuits. Despite sincere beliefs that National Service has had a wholly unsatisfactory effect upon the lives and characters of many young men in leading to excessive drinking and sexual promiscuity, it is generally accepted that it has provided the majority of those serving with challenge and adventure suitable to their age and needs. The nation, and the young men themselves, will be the poorer unless the Youth Service can provide, in relevant civilian terms, opportunities to match those which come through National Service. The cessation of National Service and the arrival of the 'bulge' will make the Youth Service responsible for about a million more young people in 1964 than it had to cater for in 1958.

A third factor which strengthened the Albemarle Committee's belief that the Youth Service needed immediate support is the unfortunate emergence of a new climate of crime and delinquency. Without wishing to exaggerate the incidence of delinquency, which still involves in any one year only about 2 per cent of boys aged fourteen and less than twenty-one and only 0·2 per cent of girls of the same age group, it is still the fact that, of the total number of persons found guilty of indictable offences in 1957, about 50 per cent were less than twenty-one, and 30 per cent were aged 14–20 inclusive. Whether the figures are large or small in relation to the total population, the crime problem is largely a youth problem, and one in which the public can reasonably expect the Youth Service to play a considerable part in its solution.

To meet the challenge of the 'bulge' and the opportunities provided by the cessation of National Service, as well as to serve the nation in helping to combat the incidence of delinquency among young people, demands a Youth Service made up of virile and enthusiastic men and women dedicated to their calling. This was scarcely the position which confronted the members of the Albemarle Committee during their investigations. Instead, the Committee found leadership suffering from shortage of money and lack of encouragement. Leaders, feeling unsupported and unappreciated, have looked for some sign that their work is nationally recognized as important. They find it neither in official expressions of policy nor in the rewards of a salary scale for those who are full-time which would put the work on a level with cognate professions. They seem to themselves to be in danger of becoming cut off from the march of social and educational advance, and it is neither surprising that the total number of full-time youth leaders is declining nor that full-time posts are failing to attract applicants with the desired qualities.

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[*] Ministry of Education. The Youth Service in England and Wales; Report of the Committee Appointed by the Minister of Education in November, 1958. Pp. iv+136. (Cmnd. 929.) (London: H.M. Stationery Office, 1960.) 6s. net.

[†] Ministry of Education. 15 to 18: A Report of the Central Advisory Council for Education (England). Vol. 1: Report. Pp. xxxi+519. (London: H.M. Stationery Office, 1959.) 12s. 6d. See also *Nature*, February 6, p. 341.

For the running down of the Youth Service and the decline in morale of its leadership, the main responsibility must be pinned on the door of the Ministry of Education. The high hopes raised in 1939 by the issue of Circular 1486 by the Board of Education, which was commended (see *Nature*, 146, 627; 1940) as an imaginative act by the Government, have been dashed by subsequent performance. During the ten years up to the end of 1958, the Ministry has not issued a single circular devoted solely to the Youth Service. During the same period there have been ten circulars having some bearing on the Youth Service and seven of them imposed restrictions. The Select Committee on Estimates, in its Report of July 1957, referred to this discouraging effect and put it down to "lack of interest on the part of the Ministry of Education in the present state of the Service and the apathy about its future"*.

In view of the Ministry discouragements, it is not surprising that many local education authorities have shown quixotic attitudes towards the Youth Service. It may seem incredible, but some so-called progressive authorities have no youth committee and no youth officer. Even authorities which value the Service show surprising variations in their practical approaches to its methods of organization and administration and, as a result, there is no accepted minimum standard of services which voluntary bodies of standing can expect from every authority as a matter of course.

It is, indeed, a tribute to the voluntary organizations—and of the voluntary principle which permeates the Youth Service—that the Youth Service has continued to remain in being at all. Apart from the time and devotion given to the Service by so many voluntary helpers—which may include members of local authorities in their official capacities as well as unpaid representatives of voluntary organizations—the taxpayers and ratepayers are usually surprised to learn that the financial weight of the Youth Service falls mainly upon the voluntary organizations. The Standing Conference of National Voluntary Youth Organizations has stated that it is not unusual for its member organizations to have to raise from private and voluntary sources 90 per cent of its total yearly expenditure on headquarters administration, the provision of regional organizers and the training of leaders. Against this, the total sum spent on the Youth Service from the Ministry of Education and local education authority funds in 1957–58 amounted to the not very grand total of £2¾ million; of every pound spent on education, about 1*d*. went to the Youth Service. Where the Ministry can fairly be charged with dereliction of duty is brought home by the Committee's finding that, during the twelve years up to and including 1957–58, direct expenditure by the Ministry on the Youth Service in terms of real money has fallen by a quarter. It is little wonder that many voluntary workers have lost heart and grown weary of working in the dingy, dilapidated buildings with insufficient equipment which have become the trade mark of so many youth clubs.

Much of the Albemarle Report is rightly given to this analysis of why the Youth Service has failed to march with the times; public opinion should see that, in the national interest, central and local government should not again fail in their duty. To give a clear and unequivocal lead, the Committee makes certain minimum recommendations for immediate action which the Government could ill afford to ignore. Sir David Eccles is to be congratulated for accepting this and for the unparalleled action of asking the House of Commons to accept, in principle, all the recommendations in the Report on the day of its publication.

The main proposal is that the Minister should initiate a ten-year development programme, divided into two stages of five years each. During the first five years advances should be made in each sector of the Youth Service to "catch up with a situation already upon us". During the second five-year period the permanent structure should be securely established. Throughout the period of development, the Minister should have at his disposal the best possible advice and help. The Committee wisely recommends the appointment of a Youth Service Development Council which will be little concerned with administration and far more with inquiry, with the sharing of ideas and the promotion of new activities; on occasions, it may serve to rejuvenate a flagging Minister. This Council has, in fact, now been formed and consists of: the Countess of Albemarle (for a period of one year); Mr. Richard Hoggart, a member of the Albemarle Committee and author of "The Uses of Literacy"; Mr. Christopher Brasher, athlete and journalist; Dr. W. Watson, sociologist from the University of Manchester; Miss M. E. Keatley, Staffordshire head mistress; Mr. Edward Lewis, chairman of Decca; Mr. Timothy Raison, journalist; Miss Ellen McCullough, National Woman Officer of the Transport and General Workers' Union; Councillor Alfred Blake, a member of the Portsmouth City Council; Mr. G. S. Bessey, director of education for Cumberland; Miss P. C. Colson, secretary of the Central Council for Physical Recreation; Mrs. Glyn Roberts, former senior mistress in a secondary modern school in Wales; Mr. Huw Wheldon, broadcaster. The Minister of Education himself will be chairman.

As an immediate step, the Report asks the Minister to put in hand arrangements for training 'professional' youth leaders, both for the emergency and the long term. The full-time youth leader force should be increased from the present total of 700 to a provisional one of 1,300 by 1966. To bring this about, an emergency training college should be opened by the autumn of 1961; Sir David Eccles has already indicated that this will be in Leicester. To anticipate legitimate criticisms about the methods of training youth leaders, a working syllabus has been devised which bears close relationship to those used for training teachers. Much here depends upon the choice of the principal, and it is to be hoped that the over-regimentation found in some of the emergency teachers' training colleges set up after the War may be avoided.

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* Seventh Report from the Select Committee on Estimates. "The Youth Employment Service and Youth Service Grants." (London: H.M. Stationery Office, 1957.)

Besides recommendations to ensure that salary scales are commensurate with the responsibilities of the job, and that money is spent in providing building and equipment which give young people who leave school at fifteen or sixteen indoor and outdoor facilities at least equal to those enjoyed by the more privileged who remain in full-time education until they reach years of maturity, the Committee makes two recommendations which deserve close attention.

The first is that the Youth Service should be available for all young people aged 14–20 inclusive. However inadvertent, the proposal to lower the statutory age limit from fifteen to fourteen is a striking comment on one of the fundamental recommendations put forward by the Crowther Committee. This was the unhappy proposal to develop county colleges on a nation-wide scale in about twenty years time. The Albemarle Committee reports how impressed it has been "by the abundance of evidence about the earlier maturity and at least superficial sophistication of many young people in the last school year at the secondary modern school. We know that they are restless and critical then and anxious to move out of the childish world into the freedoms of adolescence. We have been equally impressed by the failure of many children's organizations to hold their members at precisely this age, and by the need all clubs and movements feel to build a bridge between school and life in the world before the boy or girl has actually left school".

No better case for the county colleges has been made out, and the Albemarle Committee is to be complimented for its realistic attitude to the problems of the less intellectually privileged in their last year at school. This minor reform to the Youth Service, however, can in no way be regarded as a substitute for what still remains as the fundamental feature in building a real service of youth. County colleges will not only serve to make earlier transition between school and work and between immaturity and maturity, but will also be the rock around which all the Youth Service should be built.

The second recommendation could easily escape general notice. It is that, during the ten-year period of development, a sub-committee of the local education committee authority (not a sub-committee of a sub-committee) should be charged with responsibility for the Youth Service. The Albemarle Committee recommends that this sub-committee should have the advice of an advisory committee with full representation from voluntary organizations. Now that the Minister has signified his acceptance of the Report, he should see to it that local authorities do not evade their responsibilities for this recommendation. The youth sub-committees should be small and executive in character, and composed of men and women not directly concerned with any youth organizations but enthusiastic about their work. It should not be regarded as yet another committee for politically elected representatives to local councils. To the charge that, with authority to plan and administer the Youth Service, should go responsibility to the electors for public funds should come the reply that the youth committee would be directly responsible to the education committee in recommending expenditure of public monies. If this proposal is adopted, the Albemarle Committee may not only have made a most effective contribution to the establishment of a more virile Youth Service in Britain, but also the beginnings of a reform in local government, which has long been creaking in many joints.

The Countess of Albemarle and her committee are to be warmly praised for the promptitude with which they have discharged their task, and for putting forward a minimum set of practical proposals which the Government would have found difficult not to accept.

CHANCE AND PROVIDENCE

Chance and Providence

God's Action in a World Governed by Scientific Law. By Dr. William G. Pollard. Pp. 190. (London: Faber and Faber, Ltd., 1959.) 16*s*. net.

Dr. W. G. POLLARD is the executive director of the Oak Ridge Institute of Nuclear Studies in Tennessee and has also, since 1952, been a priest of the American Episcopal Church. In entering the Christian ministry from an already established career as a physicist, the greatest intellectual problem with which he was faced—a more difficult and decisive question than that of the existence of God—was, as he himself tells us, the problem of reconciling the empirico-causal attitude of science with belief in the providence of God. The present work is the result of eight years of thought about this question.

It soon became clear to Dr. Pollard that it would not do simply to seek for some residual domain of experience which was inaccessible to science and in which divine action might be located. This is the 'God in the gaps' expedient, which has suffered from the successive elimination of more and more of the gaps as scientific discovery has advanced. What he did, however, come to recognize was that the statistical character of scientific law, which has so markedly characterized the modern quantum theory, was in fact really basic even to classical physics, though this was unrecognized owing to the fact that the most tractable, and so the most fully developed, branches of science were those in which this character was scarcely evident. Illuminating examples are given from the realms of fluid dynamics (the problem of turbulent flow), ballistics and the early history of the solar system. (It may be remarked in passing that Dr. Pollard's argument has little in common with the argument by which, in the *British Journal of the Philosophy of Science* for 1950, Dr. K. R. Popper defended a quite different indeterminism in classical physics, an indeterminism which is better described as unpredictability.)

Dr. Pollard then goes on to stress the point that in history, as distinct from science, it is always the individual events that are significant; it is the *facts* that the Egyptians were drowned in the Red Sea and that Washington beat Cornwallis at Yorktown that have radically affected the subsequent course of the world's history, not the probabilities that those events would happen given the particular circumstances. This point is amplified by a discussion of the difference between scientific time, in which the origin of the time-coordinate is quite arbitrary, and historical time, in which the contrast between the unalterable past and the (as yet) undetermined future is fundamental, though it is also remarked that, with quantum theory, historical time has invaded the domain of science itself. It is also emphasized that miracle, as properly understood, is in no way contradictory to scientific explanation, for it should be thought of not as an interference with or a suspension of scientific law, but as a (scientifically) highly improbable correlation of natural causes, none of which violates any known scientific law: and, as regards allegedly unique events, such as the Creation, the Incarnation and the Resurrection, no question of chance or probability arises, for it is only in the case of events which are in principle indefinitely repeatable that the concept of probability can be applied. The book concludes with a discussion of freedom and providence, in which it is suggested that something similar to, though not identical with, the principle of complementarity in physics may be needed in other fields of science in order to remove the elements of historical time from the systems which are the object of scientific study, and the conception of the twofold nature of reality is further illustrated by the use of Dr. Martin Buber's distinction between 'I-it' and 'I-thou' relationships.

This is an important, if not entirely an easy book, and it is particularly to be commended for its author's thorough grasp of both the realms with which he is concerned—those of science and of theology—and not, as so often is the case in books on this subject, with only one of them, and for his determination to think the problems through to the end and not to be content with superficial or makeshift solutions. E. L. MASCALL

RADIOISOTOPES IN MEDICAL AND BIOLOGICAL RESEARCH

Radioisotopes in Scientific Research

Proceedings of the International Conference held in Paris in September 1957, under the auspices of the United Nations Educational, Scientific and Cultural Organization. Edited by Prof. R. C. Extermann. Vol. 3: Research with Radioisotopes in Human and Animal Biology and Medicine. Pp. xxi+763. Vol. 4: Research with Radioisotopes in Plant Biology and some General Problems. Pp. xxi+690. (London and New York: Pergamon Press, 1958.) 140s. net each.

ON looking through the voluminous publication on "Research with Radioisotopes in Plant Biology" (Vol. 4) and in "Human and Animal Biology and Medicine" (Vol. 3), one could argue whether it is useful to compile so much heterogeneous material from diverse fields of biology, only because the same technique is applied.

However, when reading the two volumes under discussion, these doubts are dispelled, for in the case of research with isotopes the common tie is not only a special technique but also a typical point of view. We might call this a new kind of dynamic biology, which only began with the introduction of the tracer techniques, and the results of which have nowhere been laid down more impressively than in these collective volumes.

The dynamic state of certain constituents of organisms is continuously being subjected to a kinetic analysis, regardless whether one examines the transport of electrolytes through the cell membrane and other general permeability phenomena, the effect of fertilizers, the synthesis of protein and nucleic acid, raw products of plants such as rubber, or the turnover of the most vital constituents of the healthy or pathological organism. The study of work in fields unfamiliar to the reader therefore acts as a stimulus for new ideas and the devising of new techniques in his own work.

Vol. 4, which is devoted to "Plant Biology and some General Problems", also comprises a series of specialized work in the field of animal physiology, so that zoologists and medical men for this reason should consult also both volumes.

It is very commendable that several papers are produced with many more useful details than were contained in the original lectures, and that techniques in particular are discussed at great length and many useful illustrations are added. OTTO HUG

PETROLEUM GEOLOGY

Geochemical Methods of Prospecting and Exploration for Petroleum and Natural Gas.

By A. A. Kartsev, Z. A. Tabasaranskii, M. I. Subbota and G. A. Mogilevskii. English translation edited by Paul A. Witherspoon and William D. Romey. Pp. xxiii+349. (Berkeley and Los Angeles: University of California Press; London: Cambridge University Press, 1959.) 94s. net.

THIS book presents a comprehensive survey of the geochemical methods of petroleum and natural gas prospecting and exploration being used in the U.S.S.R. in 1954 and reflects the large scale of the Russian effort in this field. The authors in a preface state that the changes which have taken place between publication of the Russian and American editions are mainly a matter of emphasis. However, it is probable that some of the analytical techniques have by now been modified or superseded and that methods which are briefly mentioned as being developed are now established techniques.

The standard of the translation is high but the lack of exact English equivalents for some of the Russian technical terms and the differences in Russian and English syntax has, on occasion, caused some difficulties. There are more typographical errors than should be expected in a technical book of this character, and the maps and figures, as in so many Russian publications, are often far from clear. In particular, no scales are given for the maps.

The first three chapters deal with the basic concept underlying the subject and include a more than adequate description of the geochemistry of oil formations and of gases. The remaining chapters consider geochemical methods which have proved to have a definite practical application. These include gas and core gas surveys, oil and gas logging, bitumen, bitumen luminescence, hydrochemical, radioactive, soil salt, oxidation reduction potential and microbiological methods. Various other methods which have been mentioned in the Russian literature, but which were not adjudged by a conference on geochemical methods of prospecting for petroleum and natural gas (Neftyanoye Khozyaistvo, 1955) to be of proved usefulness, are not considered.

Each method is described in the logical order of theoretical basis, field procedure, analytical techniques and interpretation of results. The equipment for various alternative sampling and analytical techniques is described in detail. Although, in general, more comment on the relative merits of the methods and techniques would have been appreciated, the authors do not hesitate to criticize those methods, whether Russian or American, with which they do not agree on theoretical grounds.

Mention is made on a number of occasions of the difficulties of distinguishing true anomalies from false anomalies resulting from biological activity and other causes. The interpretation of leakage dispersion patterns is dependent on a considerable number of variables, yet this aspect of the subject is only relatively briefly discussed. The inclusion of additional case histories presenting all the data, their interpretation and the results of subsequent drilling would have been of great interest.

The authors consistently emphasize the necessity for correlating the data from different types of geochemical methods and the results of these methods with geophysical and geological information. Geochemical methods of prospecting and exploration are not a universal panacea but are additional tools in the hands of the geologist. The geochemical prospecting methods described in this book are considered to have proved their worth under certain conditions but, as with all methods of prospecting, their application is limited to those areas where conditions are favourable.

Despite some few shortcomings, this book gives a well-balanced picture of the application of geochemical techniques and is recommended to petroleum geologists interested in obtaining a knowledge of these new and rapidly developing methods, which may give valuable additional information in the search for oil deposits. JOHN S. TOOMS

GROWTH OF PRIMARY EDUCATION

Primary Education

Suggestions for the Consideration of Teachers and Others concerned with the Work of Primary Schools. Pp. xiii+334. (London: H.M. Stationery Office, 1959.) 10*s*. net.

THE development of efficient methods of teaching in primary schools, improved organization of the schools themselves, and a markedly greater degree of achievement by the children have resulted from a widespread and quickening interest in primary work by teachers and the general public. This is well brought out in this Ministry of Education publication, which shows that the rise in children's achievements has not come about suddenly or by mechanical application of any special methods or practice, but because teachers, from their experience and from the growing body of research available to them, have come to understand better the ways in which children learn, and have applied their knowledge to good purpose.

The book is in effect an anthology of the ideas and practices which present-day teachers are successfully developing in the schools.

Twenty years ago junior schools were only tentatively striking out towards what is now common practice. Less than half the children between eight and twelve years of age were in schools separate from those containing older children also. Now, more than nine-tenths of school children up to eleven years of age are in separate infants schools, separate junior schools, or schools for children between five and eleven only.

'Primary Education' calls for a more critical consideration by teachers of the quality and substance of what is offered to the children for their learning, and for a firmer realization that children's capacities, whether small or great, should be exercised to the full.

For the man of science, one of the most interesting sections is that concerned with the teaching of mathematics. There is little doubt that deficiencies in the primary school must bear considerable responsibility for subsequent failures in mathematics. Teachers in primary schools can do much to prevent the positive dislike of mathematics which is so common in older children. It is to be hoped that the new three-year training college courses will produce primary school teachers who can get away from teaching mathematics by rote and will be able to teach young children in a meaningful way.

CONURBATIONS OF GREAT BRITAIN

The Conurbations of Great Britain

By T. W. Freeman. With a chapter on the Scottish Conurbations by Catherine P. Snodgrass. Pp. xii+393 (66 maps). (Manchester: The University Press, 1959.) 37*s*. 6*d*. net.

PATRICK GEDDES' term 'conurbation', more narrowly defined by Fawcett, is a vitally important but vague geographical concept that calls for searching analysis of criteria and their cartographic representation. For this reason we turn to this book with special interest.

As to content, Chaps. 1 and 12 should be read first. Then those of the intervening chapters about the conurbations (or the ensemble of minor conurbations) of most interest should be read. For the book is heavy in topographical detail that becomes irksome in spite of the aid of maps. The sectional treatment of each conurbation includes physical features, historical growth, distribution and movements of population, and, a main feature, the growth of the patterns of administrative areas. The work depends on geographical and historical studies as well as on the interesting data contained in reports of past commissioners and the census reports. It contains 66 maps, 25 of which show the changing patterns of the administrative areas in each conurbation.

Freeman, following Fawcett, has attempted to define every conurbation by the extent of its built-up area. There are seven major conurbations, each of which receives a separate chapter in the book. Fourteen minor conurbations have 250,000–750,000 inhabitants. These include several 'clusters' of special interest—the South Wales valleys, Sheffield–Rotherham (Geddes' South Riding), the Lancashire Coalfield (Wigan–St. Helens), and the Potteries. Some of these areas already form more extensive and almost contiguous clusters, as envisaged by Geddes. The whole South Wales area (Geddes' Waleston) has 1·1 million people. Nottingham and Derby are also linked with 850,000. The Sheffield and Barnsley areas are virtually one. Manchester and Liverpool are interconnected through the Lancashire coalfield area (Geddes' Lancaston). The smaller conurbations (50,000–250,000), 75 in number, are treated in two chapters. Portrayals are necessarily brief and encyclopædic.

This book is essentially a descriptive compilation. This is how Freeman chose to write it; but the approach will not advance geography as a research discipline or as a contributor to planning. He does not formulate and systematically develop any geographical concepts with regard to urban growth, an approach that would necessitate a complete reorganization of the book, but would yield more worth-while results. Geographers need to concentrate on particular problems in the spatial structure of our urban society, some of which are sandwiched in the pages of this book. Here are involved questions of criteria, cartographic analysis, and interpretation that cry out for investigation, and are especially fitted to the expertize of the geographer. The defects of this book point to this need, and it is to be hoped that Freeman and his colleagues will apply themselves to it.

R. E. DICKINSON

STUDIES IN RHEOLOGY

Transactions of the Society of Rheology

Vol. 2. Edited by R. D. Andrews. Pp. ii+314. (New York: Interscience Publishers, Inc.; London: Interscience Publishers, Ltd., 1959.) 64*s*.

Rheology of Disperse Systems

Proceedings of a Conference organized by the British Society of Rheology, and held at the University College of Swansea, September 1957. Edited by C. C. Mill. Pp. vii + 223. (London and New York: Pergamon Press, 1959.) 60*s*. net.

THE "Transactions of the Society of Rheology" constitute the proceedings of the annual conferences of the American Society. In 1957, the conference was held in the Textile Research Institute, Princeton, New Jersey. The second of these volumes contains sixteen complete papers and a number of abstracts.

The organizers are to be congratulated on the excellent balance of the programme. It would be tempting to give a heavy preponderance to papers dealing with work in the one or two industries which now employ a high proportion of rheologists; but this has not been done. Theory and practice over a wide field are discussed.

In an introductory paper, A. S. Nowick outlines the career of C. M. Zener, who received the Bingham Medal. Dr. Zener is distinguished in other fields, and it is estimated that his rheological work constitutes less than half his scientific output.

On the more academic side, the subjects treated include work on viscosity–concentration relations in suspensions, relaxation spectra distributions of high polymers, equations for the interconversion of relaxation and retardation spectra and a general study of capillary viscometry.

Papers on applied rheology range over a wide field: the peeling of adhesive tapes, liquid flow in roll nips, creep of snow, mechanical breakdown of bundles of fibres and a fair number of articles on high polymers.

The paper by A. B. Metzner and M. Whitlock is worthy of special mention. Since Osborne Reynolds published his classical work, it has been well known that certain pastes and suspensions become firmer on shearing. All such phenomena have tended to be classed as 'dilatancy', it being assumed that closely packed particles must first pass into open packing before the system can flow, thus producing an increase in volume. This paper confirms what has long been suspected, that the increase in consistency is not necessarily associated with dilatancy at all and often happens at concentrations not corresponding to close packing. The true explanation remains an outstanding problem for rheologists.

The 'Transactions' will certainly provide an excellent annual survey of the progress of rheology in the United States. It is a pity only that this particular volume has taken so long to appear.

It is difficult to review a collection of eleven papers which, though they are all concerned with dispersed systems (must one omit the grammatically required 's' ?), cover so wide a range of subjects and materials. Yet it is fair to say that, though the standard of the articles differs considerably, the general level is remarkably high.

Prof. J. G. Oldroyd's presidential address deals with the mathematics of 'complicated rheological properties', because, as he points out, simple properties are of little interest to the mathematician. This is fortunate, in view of the complexity of the materials generally studied by practical rheologists.

S. G. Mason and W. Bartok contribute a paper of considerable theoretical interest on the hydrodynamics of suspensions and emulsions, and R. L. Whitmore gives some reasons why, on theoretical grounds, suspensions flowing through capillaries fail to obey Poiseuille's fourth-power law, applying his theory to the very important problems of the flow of blood. Since this paper was read, it has stimulated considerable further work. Also N. Casson's and E. Bantoft's contributions, in which they propose a very simple but hitherto unused equation to relate shear stress to rate of shear, has been followed by the publication of descriptions of a number of applications to materials other than varnishes and inks, including blood and molten chocolate. The latter forms the subject of an article by E. H. Steiner, but it was after the conference that he discovered the usefulness of Casson's equation for this material.

A. Jobling and J. E. Roberts, using Weissenberg's deformable membrane technique, describe complex dilatancy effects in certain suspensions which still elude any mechanical explanation and yield a strong incentive to further study. Other papers deal with high polymers, adhesive sealing compositions ('lutings'), starch-clay mixtures and gaseous fluidized beds. This is clearly a conference which immediately activated further research, thus fulfilling the prime purpose of all such gatherings.

The publishers are to be congratulated on the excellent production of the book. A welcome appendix gives the addresses of all authors and contributors to the discussions.

G. W. SCOTT BLAIR

TERCENTENARY CELEBRATIONS OF THE ROYAL SOCIETY

THE programme of events which are to comprise the tercentenary celebrations of the Royal Society, due to take place during July 18–26, indicates that not only is the historical importance of the event to be appropriately marked but also that those who are to take part will have a good opportunity to become acquainted with active research work now being carried out in Great Britain.

Formal participation in the full programme will be limited to Fellows and Foreign Members of the Society and to those already invited to represent at the celebrations national academies of science, selected universities throughout the world, and appropriate international scientific organizations. The majority of the Foreign Members of the Society have already notified their intention of being present, and other distinguished men of science are among those who are coming from some fifty different countries. The British Commonwealth will be particularly well represented by large numbers, many of whom are Fellows of the Royal Society. The presence in London of so many leading scientists promises to make the occasion an outstanding scientific event.

In addition to the formal participants and their accompanying relatives, for whom the full programme of events has been principally devised, there will be a number of events to which others will be invited, notably the formal opening ceremony on the afternoon of July 19, which is to take place in the Royal Albert Hall. The ceremony will take the form of a few short speeches and the presentation of formal congratulatory messages by selected representatives attending the celebrations. For the occasion, Sir Arthur Bliss, Master of the Queen's Musick, is composing a "Salute to the Royal Society", and this will be played by Dr. G. Thalben-Ball on the organ together with the trumpeters and other musicians from the Royal Military School of Music, Kneller Hall. After an interval for tea the Society's president, Sir Cyril Hinshelwood, will deliver the tercentenary address.

In honour of the occasion the Universities of Oxford, Cambridge and London will have special honorary degree ceremonies, when degrees will be conferred on distinguished scientists attending the celebrations.

The Oxford and Cambridge ceremonies will take place on July 21 and July 25, respectively, during visits to these Universities. While in Oxford, visits will be paid to the Colleges which will receive the Royal Society visitors at lunch. The conferment of honorary degrees will take place in the Sheldonian Theatre, the architect of which was Sir Christopher Wren, former president of the Royal Society, and this will be followed by a garden party at Wadham College, where meetings of importance in relation to the foundation of the Society took place. John Wilkins, Bishop of Chester and Warden of Wadham, was one of the Society's first secretaries. The visit to Cambridge will have a similar pattern, and in both Universities special exhibits relating to former Fellows of the Society are being specially arranged.

The University of London ceremony will take place in the Royal Festival Hall on July 22, where H.M. Queen Elizabeth the Queen Mother as Chancellor of the University will confer honorary degrees.

The following lectures will be delivered during the period of the celebrations in the Royal Institution, the Royal College of Surgeons, and the Beveridge Hall of the University of London: "Genetics and the Structure of Biology", by Prof. C. D. Darlington; "Trends in Aeronautical Science and Engineering", by Sir Arnold Hall; "The Evolution of Nuclear Power Plant Design", by Sir Christopher Hinton; "The Physics and Chemistry of Nervous Conduction", by Prof. A. L. Hodgkin; "Molecules in Crystals", by Dr. Dorothy Hodgkin; "The Investigation of the Universe by Radio Astronomy", by Prof. A. C. B. Lovell; "The Problems of Transplantation", by Prof. P. B. Medawar; "The Study of Nuclear Interactions at Very Great Energies", by Prof. C. F. Powell; "New Horizons in Organic Chemistry", by Sir Alexander Todd; "The Metamorphosis of Insects", by Prof. V. B. Wigglesworth.

The programme includes visits to university, government and industrial research laboratories which are or have been associated with the work of Fellows of the Society. In addition to visits to the Universities of Oxford and Cambridge, there are formally organized visits to the Imperial College of Science and Technology and University College in London, as well as informal visits privately arranged. The National Physical Laboratory, the Royal Botanic Gardens, Kew, the National Institutes for Medical Research, for Research in Nuclear Science and for Research in Dairying, as well as the Royal Aircraft Establishment, the Atomic Energy Research Establishment, the Geological Survey and Museum, and the Radio Research Station and Road Research Laboratory (Department of Scientific and Industrial Research) are among the national research institutions to be visited. The British Museum (Natural History) and the Science Museum are arranging special visits. The Associated Electrical Industries, Ltd., Research Laboratory at Aldermaston, the British Petroleum Research Centre, the General Electric Co. Research Laboratories, the Glaxo Research Laboratories, Imperial Chemical Industries, Ltd. (Plastics Division), the Wellcome Research Laboratories, and several research association laboratories provide a quota of industrial research visits.

Other events or visits recall the historical associations of the Society. The foundation of the Society in Gresham College near Guildhall in the City of London will be commemorated not only by two City receptions, one by the Lord Mayor in Guildhall and the other by the twelve great City Companies in the Mercers' Hall, but also on Sunday, July 24, by a sermon relating to Sir Christopher Wren, to be preached in St. Paul's Cathedral, of which he was the architect. One of the tours will re-visit the present-day Gresham College and also the other former homes of the Society in Crane Court off Fleet Street and Somerset House. Visits to Greenwich, where King Charles II instituted the Royal Greenwich Observatory, and the Chelsea Royal Hospital, built by Wren on a site once briefly owned by the Royal Society, are direct links with the Society's Royal founder. The Royal Mint, where Newton presided as master for many years, and Newton's birthplace at Woolsthorpe are also included in the places to be visited.

The Shell International Petroleum Company is making a special film for the occasion. This will be shown in the Royal Festival Hall on July 22, in a programme which will also include reference to the value of the film as a tool in scientific research. At the Society's tercentenary conversazione, to be held in Burlington House on July 23, a series of exhibits giving a presentation of current research work will be included in the programme of the evening, during which there will also be on display the congratulatory addresses received by the Society on the occasion of its three hundredth birthday. Some of the Society's treasures, such as the original reflecting telescope made by Sir Isaac Newton, the manuscript of Newton's "Principia", the Society's Mace and the Charter Book, both of which are gifts from the Society's Royal founder, will also be displayed on this occasion.

The celebrations will conclude with the tercentenary banquet on July 26, when the toast of "The Royal Society" will be proposed by the Prime Minister.

The issue of the first volumes of "The Correspondence of Isaac Newton", edited by Prof. H. W. Turnbull, happily coincides with the tercentenary year of the Society as a tribute to the Society's greatest Fellow. Two further historical works are being specially issued to mark the tercentenary occasion. These are a new brief history of the Society by Prof. E. N. da C. Andrade, and a volume of the Society's *Notes and Records* which is edited by Sir Harold Hartley. This special volume will give an account of the founder Fellows including Robert Boyle, Christopher Wren, Sir Robert Moray, Viscount Brouncker, Robert Hooke and others, as well as articles on the origins of the Society.

D. C. MARTIN

SOME CONSEQUENCES OF EXPANSION OF THE EARTH

By PROF. J. TUZO WILSON

Department of Geophysics, University of Toronto

IN 1937 Dirac[1] suggested that the gravitational constant G is decreasing with time, proportionally to t^{-1}, where t is the age of the universe. At that time t was considered from astronomical evidence to be about 2×10^9 years, although age determinations indicated a greater age.

In two recent papers Dicke[2,3] has discussed some of the consequences of this hypothesis using more recent estimates for t of $6\cdot5 \times 10^9$ and 12×10^9 years, respectively. Using the first figure he concludes that in the past $3\cdot25 \times 10^9$ years the circumference of the Earth would have increased by about 4·5 per cent or 1,100 miles at an average rate of about 0·5 mm. a year. He suggests that this expansion should still be recorded in cracks widely distributed over the ocean floors. He also finds no compelling evidence of a physical nature for or against Dirac's hypothesis and speculates whether "the history of the solar system as seen through the eyes of astronomers and geologists may enable fundamental conclusions to be drawn regarding physical interactions in the past". Egyed[4] is another who has long advocated an expansion of the Earth on geological grounds and has estimated a rate of increase in circumference of 3·8 mm. a year.

It is proposed here to examine the geological and geophysical evidence briefly in the light of recent observations.

The information about the Earth has been chiefly contributed by stratigraphers, Precambrian geologists, students of physics of the Earth and oceanographers studying the sea-floor. Unfortunately, it has never been well integrated.

As a result of the International Geophysical Year, progress in oceanography has recently been very rapid. In 1956 Ewing and Heezen[5] suggested that the mid-ocean ridges form a continuous, seismically active belt, extending through the Atlantic, Indian and South Pacific Oceans. They[6] have since stated that a median rift valley already noticed by Hill[7] is a characteristic feature, that it may be the locus of the earthquake foci, and that it (and perhaps the ridge in general) have intense magnetic anomalies "due to a subsurface body of high magnetic susceptibility". A branch of this rift has been traced to the Gulf of Aden[8] and thence to the rift valleys of Africa[9], which have similar geophysical features.

Heezen[10] suggests that the African rifts represent the start of another ocean and that existing ocean basins may have grown by the widening of such rifts under tension, "if, as the Earth's exterior expanded, the continental crust maintained a relatively constant area". Since the oceans occupy more than two-thirds of the Earth's surface, such an expansion would involve nearly doubling the Earth's radius during geological time. The impossibility of this has been pointed out by Teller[11] and is appreciated by Heezen, who mentions that the postulate of an expanding Earth "may have other more serious objection of an astronomical nature".

If, on the other hand, one postulates that only the mid-ocean ridges and not the whole ocean basins have grown, the expansion would have been much less. Their width varies and is not accurately known, but Heezen's estimates seem to range from 400 to 800 miles. The length of the active ridges is 40,000 miles; but echo-soundings by U.S. Submarine *Nautilus* suggest a continuation of the mid-Atlantic ridge across the Arctic basin, parallel with the Lomonosov ridge[12], and it has been suggested that some mid-Pacific ridges form part of the system, although seismically quiescent at present[13,14]. Including these gives a minimum length of the ridges of 60,000 miles and a minimum superficial expansion of about $2\cdot4 \times 10^7$ square miles. This would require an increase in the Earth's circumference of about 6 per cent, and the formation of the ridges has probably required all geological time because there are no important abandoned ridges or any obvious way of disguising them. This is close to Dicke's estimate. This does not prove that expansion of the Earth has occurred or if it has that it is due to change in G, but it is worth considering some of the consequences of accepting that hypothesis.

Bullard[15], who devised a method of measuring heat flow through the ocean floor, and Von Herzen[16] have remarked that over most parts of the ocean floor the flow of heat is equal to that on continents but that it is higher over the mid-ocean ridges. This would be explained if horizontal separation of the original crust along the ridges continually exposed hotter interior layers. This high heat-flow and the absence of any earthquakes of focal depth greater than 70 km. along the ridges suggest that the Earth is only brittle to stresses of short duration to a depth of 70 km. beneath the ridges.

The lower temperature gradients and the occurrence of earthquakes at depths of up to 700 km. under other parts of ocean floors and continents suggest that those parts may be brittle to 700 km.

The alternative explanation that rising convection currents cause the high heat flows is as satisfactory for the mid-ocean ridges, but it has never been shown how sinking currents could account for the very varying types of coasts of different oceans or the patterns of mountain belts. Reduced heat flows found in some trenches could be due to recent flow into them of cold turbidity currents and submarine oozes.

Since the distance to the zone of flow under the mid-ocean ridges may be only one-tenth as much as it is elsewhere, we might expect a rift once formed to remain the site of further expansion.

Opening of rifts along the mid-ocean ridges would tend to make the rest of the Earth's surface buckle at a distance from them. That is where the continents with their mountain system are found. There the increase in radius of curvature would produce local compression above a level of no strain (or neutral surface) along with tension below it. A similar general situation was postulated by Jeffreys[17] as a result of contraction in the Earth, and it was used by Scheidegger[18,19] and Wilson[14,20] to explain the details of mountains, island arcs, grabens and other features. Their explanations are still applicable in principle to the new hypothesis. At the same time, since the brittle shells on the Earth's surface are spherical not cylindrical, the buckling which is considered to produce mountain building would shift in direction as well as in position from time to time. It is conceivable that this change every few hundred million years might be as large as a right angle (for example, from Ural or Appalachian directions to Himalayan or Cordilleran directions), while minor adjustments and seismicity might be expected everywhere.

The view that continents are zoned and have grown during geological time over parts of the former ocean floors[14,21] is compatible with this hypothesis. Indeed expansion removes a major obstacle to this theory. If continents had grown on a shrinking Earth, the oceans would have flooded the continents, and this has not happened.

Recent work[22] on the isotopic ratio of lead ores having volcanic associations also supports the view that they (and hence by analogy other volcanic rocks) have not been reworked but are being emplaced directly from the mantle. This supports the opinion that continents are growing and not being reworked.

The observation that continents are spaced as far from mid-ocean ridges as possible and that there is some parallelism between their margins could be a consequence of the conditions necessary to produce buckling.

The view that continents have drifted apart is supported to a small degree but not to the extent desired by some advocates of continental drift and some students of palæomagnetism. Nevertheless, expansion along the ridges would alter the Earth's principal moments of inertia and could thus induce polar wandering.

One of the chief arguments used in support of convection currents has been that they might explain the rapid horizontal motions observed along large faults such as the San Andreas in California, and the horizontal first motion observed in circum-Pacific earthquakes, although the manner in which they could do this has not been explained. But such motions are also a consequence of the present hypothesis. An active mid-ocean ridge extends across the South Pacific Ocean from the vicinity of New Zealand to South America, where it terminates in two or three branches against the Andes. If this ridge is widening at the rate of about 0·5 mm. a year and if along the Andes and Cordillera there is an existing line of fracture due to buckling, then a northward shearing motion of the Pacific Ocean relative to the Americas could take place along the mountains north of the ridge. Such a northward motion of the Pacific has long been postulated, and the rate is perhaps compatible with the rather uncertain geological evidence[23].

That this shear motion would not occur south of Chile may perhaps explain the absence of any equivalent to the Isthmus of Panama across Drake Passage. Similar motions in the past could account for other shifts, such as those along the Great Glen fault[24] or the Murray scarp[25].

Isostatic balance would certainly be effective in the model Earth proposed, but adjustment would be more readily achieved for the Earth as a whole and for sea-mounts over mid-ocean ridges than elsewhere. This appears to be supported by observations. The suggestions from studies of Earth satellites that the Earth is somewhat pear-shaped[26] might result from the wider distribution of mid-ocean ridges in the southern hemisphere; but it appears to be more easily explained by the recent removal of ice-sheets. Contrary to previous opinion, Sacks and Strelkov[27] report that large ice sheets covered much of northern Siberia during the Pleistocene. Thus the geoid is still much depressed in a zone centred along latitude 60° N., and G. Pratt[28] has reported depression over Antarctica. This would produce a pear-shape without implying great strength over periods of long duration.

It is perhaps worth noting that to a first approximation the mid-ocean ridges lie in the shape of a reversed J folded about the Earth, the stem being in the Atlantic and hook being in the Southern Ocean. Two branches extend northward into the Indian and Pacific Oceans. Also to a first approximation the continents lie in a similar but complementary pattern of a J inverted and folded about the Earth with the stem through Antarctica and the Americas and the hook through Eurasia. Two branches extend southwards into Africa and Australia.

Perhaps this was the initial fracture pattern of the Earth; but the two parts have developed differently. Whereas the mid-ocean ridges have parted without migrating and have emitted basalts, the continental fractures have buckled without parting and have migrated, moving outwards about their nuclei which seem to preserve evidence of a different, more volcanic and perhaps hotter history of the Earth during a period 2,000–3,000 million years ago[30,31]. It is only the average position of the continental system that at all resembles a *J* for to-day the active fractures have the shape of a *T*, folded about the Earth with its junction in Indonesia, but there are links between the two systems, for example, between Gibraltar and the Azores, between New Zealand and the ridge to the south and in the scarps from California to Hawaii. The volcanoes of the continental system emit more acid lavas than basalts.

Thus the foregoing hypothesis has the merit of appearing to explain many features of the Earth's surface, though this does not constitute a proof. Even if true, expansion at the rate here postulated could conceivably be due to phase changes[19] in the Earth's interior or perhaps to differentiation[30] of the core and mantle, but a decrease in *G* remains an inviting idea. If so a number of geophysical effects discussed by Dicke must be taken into account in calculations about the Earth's interior and these might include secondary effects due to phase changes.

I am indebted to colleagues in the Departments of Physics at the University of British Columbia and Toronto for suggestions and particularly to Profs. G. M. Volkoff and W. H. Watson.

[1] Dirac, P. A. M., *Nature*, 139, 323 (1937).

[2] Dicke, R. H., *Rev. Mod. Phys.*, 29, 355 (1957).

[3] Dicke, R. H., *Amer. Scientist*, 47, 25 (1959).

[4] Egyed, L., *Geol. Rundschau*, 46, 101 (1957).

[5] Ewing, M., and Heezen, G. C., Amer. Geophys. Union Monog., 1, 75 (1956).

[6] Ewing, M., Hirshman, J., and Heezen, B. C., Internat. Oceanog. Congr. Preprints, 24 (1959).

[7] Hill, M. N., *Deep-Sea Res.*, 3, 229 (1956).

[8] Nafe, J. E., Hennion, J. F., and Peter, G., Internat. Oceanog. Cong. Preprints, 42 (1959).

[9] Girdler, R. W., *Quart. J. Geol. Soc. Lond.*, 114, 79 (1958).

[10] Heezen, B. C., Internat. Oceanog. Cong. Preprints, 26 (1959).

[11] Teller, E., *Phys. Rev.*, 73, 801 (1948).

[12] Dietz, R. A., First Internat. Symp. Arctic Geol., Calgary, Alberta. Abstracts (1960).

[13] Menard, H. W., *Bull. Amer. Assoc. Petroleum Geol.*, 40, 2195 (1956).

[14] Wilson, J. T., *Amer. Scientist*, 47, 1 (1959).

[15] Bullard, E. C., Maxwell, A. E., and Revelle, R., "Adv. in Geophys." 3, 153 (1956).

[16] Von Herzen, R., *Nature*, 183, 882 (1959).

[17] Jeffreys, H., "The Earth", second edit., chap. 15 (1929).

[18] Scheidegger, A. E., and Wilson, J. T , *Proc. Geol. Assoc. Canada*, 3, 167 (1950).

[19] Scheidegger, A. E., *Canad. J. Phys.*, 31, 1148 (1953).

[20] Jacobs, J. A., Russell, R. D., and Wilson, J. T., "Physics and Geology" (McGraw-Hill, New York, 1959).

[21] Farquhar, R. M., Russell, R. D., and Wilson, J. T., "Handbuch der Physik", 47, 288 (1955).

[22] Stanton, R. L., and Russell, R. D., *Economic Geol.*, 54, 588 (1959).

[23] Hill, M. L., and Dibblee, jun., T. W., *Bull. Geol. Soc. Amer.*, 64, 443 (1953).

[24] Kennedy, W. Q., *Quart. J. Geol. Soc. Lond.*, 102, 41 (1946).

[25] Vacquier, V., *Nature*, 183, 452 (1959).

[26] O'Keefe, J. A., Eckels, A., and Squires, R. K., *Science*, 129, 566 (1959).

[27] Sacks, V N., and Strelkov, S. A., First Internat. Symp. Arctic Geol., Calgary, Alberta, Abstracts (1960).

[28] Pratt, G., *Geol. J.*, 125, 354 (1959).

[29] MacDonald, G. J. F., and Knopoff, L., *Geophys. J.*, 1, 284 (1958).

[30] Urey, H. C., "The Planets" (Yale University Press, 1952).

EXPERIMENTAL TRANSFORMATION OF CELLS*

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THE term 'transformation' will be used rather loosely to cover a wide range of changes produced by various experimental conditions in the structure and functional activities of cells; merely toxic effects leading to degeneration have been excluded. Most of the examples presented have been chosen from experiments on tissues and cells grown in culture, because I myself am familiar with this field.

Part of the work to be described relates to the normal differentiation of tissues, and has been included because it sheds light on some of the morphogenic principles that are concerned in certain types of cellular transformation.

Tissue Interaction

It is now known that the different types of cells of which an organ is composed may exert a profound influence on one another's structure and physiology, especially in embryonic life.

In Spemann's classical experiments on induction it was shown that in early newt embryos certain tissues act as organizers in embryonic development and induce and direct the differentiation of neighbouring structures. That this phenomenon of induction also occurs in warm-blooded animals was first demonstrated by Prof. C. H. Waddington[1] in early chick embryos explanted in culture.

Induction is not restricted to the initial stages of development; an interesting example of its operation at a later stage of embryonic life is provided by the elegant experiments on the formation of feather rudiments, recently published by Dr. P. Sengel[2] from Prof. Etienne Wolff's laboratory. The object of this work was to investigate the interaction of the dermis and epidermis during the development of the feather papillæ. First, Sengel cultivated dorsal skin from chick embryos of 6–6½ days incubation on a very simple medium; he found that under these conditions feather germs would not develop in an explant in which no feather germs had yet appeared (stage 0) unless it were associated with another tissue—neural tube, muscle, etc.; but feather rudiments formed readily in slightly older (stage 1 and stage 2) skin explants. He concluded that at stage 0, feathers would not develop in the absence of certain non-specific external factors which were unnecessary at a later stage. Next, Sengel separated the epidermis from the dermis by Moscona's method of mild tryptic digestion, and then combined stage 0 epidermis with stage 1 dermis: feather papillæ formed in the double explants. No feathers appeared, however, when stage 0 epidermis was associated with slightly older, stage 2 dermis. From this it was concluded that at stage 1 the dermis can induce feather-formation, but that by stage 2 it has lost this inductive capacity. Sengel then combined stage 2 epidermis with stage 0 dermis: feather germs developed; this result showed that by stage 2 the epidermis had acquired the power to induce the early (stage 0) dermis to form feather papillæ.

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These results are a nice example of the reciprocal influence of mesoderm and epithelium during normal development. When isolated epithelium is associated with the wrong mesoderm, however, its differentiation may be profoundly disturbed. Thus Sengel[2] found that when the dorsal epidermis from a stage 1 (6-day) embryo was associated with the tarso-metatarsal dermis of a 12-day embryo, scales developed and the grafted epidermis became transformed into the thick, precociously keratinized epithelium normally characteristic of the shank region.

My former colleague, Dr. C. B. McLoughlin (unpublished work), investigated the influence of mesenchyme from different organs on the differentiation in culture of epidermis isolated by tryptic digestion from the limb-buds of 5-day chick embryos. In control experiments isolated epidermis when combined with limb-bud mesoderm formed an islet of keratinizing squamous epithelium, but when associated with the mesoderm of the gizzard, it failed to keratinize and formed a cuboidal epithelium which sometimes contained ciliated cells and even secreted a little mucin. When enclosed by heart muscle, the epidermis again failed to keratinize and formed a cyst of extremely flattened cells.

The interaction of epithelium and connective tissue in more complex organs has been studied by Grobstein. One set of experiments concerned the submandibular gland rudiment of 13-day mouse embryos[3]. Grobstein separated the epithelial component of the gland from the mesodermal rudiment of its capsule. He found that when the isolated epithelium alone was explanted, it grew as an undifferentiated sheet of cells; when combined with mesoderm from other parts of the body it formed an inactive, rounded mass; but when re-combined with its own capsular mesoderm it formed a branching, glandular tree. Grobstein obtained exactly the same results when he interposed a porous filter between the epithelium and the mesenchyme, although the pores of the filter were too small to allow cells to pass through. From this it was clear that the glandular epithelium requires substances produced by its own capsular cells to allow it to differentiate normally, but that direct contact with the mesenchyme is not essential.

Chemical Influences

This brings us to the subject of transformations induced experimentally by chemical means.

The best known and most widely investigated example of a chemical compound that drastically affects differentiation is vitamin A. In A-deficient animals, many mucous and glandular epithelia acquire a squamous, keratinizing structure very similar to that of the normal epidermis. A similar histological change (unpublished) was recently produced by my colleague, Dr. I. Lasnitzki, who found that when the prostate glands of young mice were grown for 10 days in a protein-free, chemically defined medium containing neither vitamins nor hormones, they developed large areas of heavily keratinized, squamous epithelium which was often continuous with apparently normal secretory epithelium. It is possible, but not yet certain, that this change may be due to the absence of vitamin A in the culture medium.

The late Sir Edward Mellanby and I found that the reverse effect to that of A-deficiency could be produced by growing embryonic chicken skin in a medium containing excess vitamin A (ref. 4). In normal medium the epithelium develops into a squamous, keratinizing epidermis, but in the presence of excess vitamin A it is changed into a mucus-secreting epithelium which is sometimes ciliated.

Dr. McLoughlin (unpublished work) has shown that this response of the epidermis to vitamin A is due to a direct action on the epithelium and is not mediated by the dermis. Pure cultures of epidermis isolated from 5-day chick embryos became keratinized in normal medium, but in the presence of vitamin A it failed to become keratinized and secreted some mucin.

Dr. Stephen Pelc and I[5] investigated some of the early histochemical changes that accompany this mucous transformation, by studying the uptake of various labelled compounds by means of autoradiography. We found that of the amino-acids, leucine and methionine were taken up equally well by the epidermis in the presence or absence of added vitamin A; but the vitamin reduced the incorporation of tyrosine in the superficial cells, and greatly diminished the uptake of cystine throughout the epithelium; these effects are probably correlated with the inhibition of keratinization. The uptake of inorganic sulphate was studied since in animals it is incorporated into mucopolysaccharides but not into amino-acids. In the deeper layers of the epithelium there was little difference in uptake between the A-treated and control explants, but in the superficial cells incorporation was very slight in the keratinizing controls but rapidly increased in the A-treated skin as secretory activity developed. This suggests that in the normal skin the basal cells have a mechanism for synthesizing mucopolysaccharides, that this is blocked by keratinization but that it continues to act when keratinization is stopped by excess vitamin A. It was interesting that whereas in the controls the uptake of tritium-labelled thymidine was restricted almost entirely to the basal nuclei, in the A-treated epidermis labelled nuclei occurred at all levels. It is unlikely that any of these histochemical changes represent the fundamental action of vitamin A on the epidermis.

Another group of chemical compounds that readily produce metaplastic changes *in vivo* are the carcinogens.

Spontaneous Transformation in Cell Cultures

Finally, a brief reference will be made to a rather different type of transformation that is well known in cell culture work. To prepare cell cultures, tissues are dissociated by enzymatic digestion, and the liberated cells are grown in flasks, tubes or bottles in an undifferentiated, very actively multiplying state, sometimes for many years. In order to survive indefinitely under these very unnatural conditions, the cells adapt themselves in various ways, so that usually they come to differ widely in appearance, behaviour and metabolism from cells of the same histological type freshly isolated from the body.

An interesting and important feature of these so-called transformations in long-term cell cultures is that they sometimes end in malignancy. Dr. George Gey[6] maintains two strains of rat fibroblasts derived from the same explant: one produces malignant tumours when implanted into rats, the other does not. Dr. Sanford and her colleagues, working in Dr. W. Earle's laboratory, have two strains of cells, both descended from the same single mouse fibroblast[7]. After 1½–2 years in culture, one of these sister lines, called the 'high' strain, when grafted into mice produced lethal sarcomas in 97 per cent of the animals, while the other, termed the 'low' strain, produced sarcomas in only 1 per cent of the mice. The two strains differ in habit of growth and in metabolism, that of the 'high' strain being largely anaerobic and that of the 'low' strain almost normal, yet their antigenic properties are the same and either can immunize against the other.

Conclusions

How far are we justified in using the term 'cellular transformation' ? Tissues certainly are transformed, but are the individual cells ?

I know of no example of a structurally differentiated cell being directly changed into one of a different histological type. In the early embryo, a wide spectrum of developmental changes can be experimentally induced in the tissues, because all the component cells are at a very early stage of differentiation. When older tissues are subjected to morphogenic influences, the fully differentiated cells are not transformed but usually degenerate and are shed. The metaplastic change in the tissue is due to the fact that the structurally undifferentiated germinative cells are potentially capable of following more than one developmental path ; which path they choose depends on their threshold of response to the particular environmental conditions to which they are exposed. In the normal animal, a striking instance of the germinative cells of an epithelium switching alternately from one developmental pathway t another is provided by the vagina : during diœstru the basal cells differentiate into mucous elements but when exposed to the influence of œstroge they form a squamous, keratinizing epithelium instead and the mucous cells are shed.

As Dr. Paul Weiss[8] has pointed out, the germinativ cells of older tissues are not really undifferentiated since their developmental repertory, though wider tha is sometimes realized, is nevertheless extremel limited as compared with that of early embryoni tissues.

Personally, I do not think that we can conside this switching of germinative cells from one develop mental pathway to another as a transformation i the strict sense of the word. As regards the type o 'transformation' that occurs in unorganized cel cultures, it is still uncertain whether the changes tha appear in the cultures represent a real transformatio of the cell population as a whole, or whether the are due to the selection of variant cells that ar peculiarly well adapted for growth in this grossl abnormal environment. I think that sometime at least, selection is the more probable explanatio of the phenomena observed.

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[1] Waddington, C. H., *Phil. Trans. Roy. Soc.*, **221**, 170 (1931).

[2] Sengel, P., *Ann. Biol.*, **34**, 29 (1958).

[3] Grobstein, C., *J. Exp. Zool.*, **124**, 383 (1953).

[4] Fell, H. B , and Mellanby, E., *J. Physiol.*, **119**, 470 (1953). Fel H. B., *Proc. Roy Soc.*, B, **146**, 242 (1957).

[5] Pelc, S. F., and Fell, H. B., *Exp. Cell. Res.*, **19**, 99 (1960).

[6] Gey, G., *J. Nat. Cancer Inst.*, 19, 784 (1956).

[7] Earle, W., *J. Physiol.*, **119**, 781 (1956).

[8] Weiss, P., *Lab. Invest.*, **8**, 415 (1959).

FOURAH BAY COLLEGE: UNIVERSITY COLLEGE OF SIERRA LEONE

By Dr. F. J. TAYLOR
Head of the Department of Botany
AND
Dr. E. DOWNING
Head of the Department of Chemistry

ON January 28, His Excellency Sir Maurice H. Dorman, governor of Sierra Leone and deputy visitor of the College, presented the Royal Charter to Fourah Bay College, incorporating it as the University College of Sierra Leone. Thus was inaugurated a new phase in the development of the College, although it has functioned as a university college for more than eighty years.

Fourah Bay College itself was founded in Freetown in 1827 by the Church Missionary Society. Before that, in 1816, a precursor of the College, the 'Church Institute', had been founded by the Church Missionary Society in the village of Leicester, just outside Freetown. This became a 'Seminary' in 1819 and moved to another village, Regent, in 1820. Its main function, and that of the early Fourah Bay College, was the training of evangelists in the belief that any continuous evangelistic work in West Africa should be undertaken by Africans.

The ambitiousness of the establishment of the College may be gauged from the fact that although a township with European traders had been establishe on the site of Freetown in the fifteenth century, th Colony of Sierra Leone was only established in 180 with a population mainly of freed slaves who wer brought from Nova Scotia in 1792 (they had fough for the British in the American War of Independenc and were dissatisfied with their rewards of grants o land in Nova Scotia) and from Jamaica in 1802. I the following years the population was increased by large number of Africans liberated from capture slave ships. Most of these were not acceptable by th more cultured repatriates from the United State and were established in villages along the coast of th Colony and in the hills above Freetown.

In 1827 the College was established with si students in a house that had at one time housed slave dealer. Though numbers were small, th students soon achieved prominence in West Africa affairs. One of the first students was Samuel Crowthe who qualified, became a teacher for some years, an then returned to the College as a tutor. In 1841 h and the Rev. J. F. Schon led the first Niger Expedition, and on his return he went to England, where he was ordained in 1843. He returned to Sierra Leone, and then proceeded to his native Nigeria, where he pioneered the spread of Christianity, and was consecrated Bishop of Lagos in 1864—the first African bishop of modern times.

During the Victorian era, the College was cosmopolitan, drawing its tutors from England, Africa, the United States and Germany; the first Principal was a Swiss. But teaching was not the sole occupation of the staff. Grammars, dictionaries and translations of the African languages came out of Fourah Bay. Of these, Koelle's "Polyglotta Africana" (1854), a comparative study of a great number of West African languages, is still a basic work.

By the early 1840's the number of students was more than thirty and the accommodation too cramped. A new building was initiated and was completed in 1848. This was sited on the isthmus between Fourah Bay and Destruction Bay (where the captured slave ships were broken up). This building, which still stands, housed the College for nearly a hundred years.

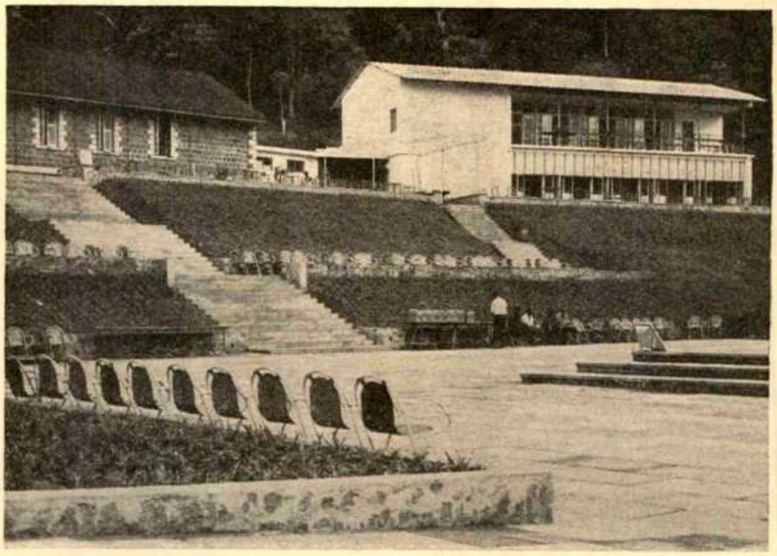

Fig. 1. The Students' Union. On the left is one of the original buildings of the hospital; on the right part of the new Union building. In the foreground is the Assembly Arena (Photo: T. Lloyd)

In 1870, the Rev. R. Sunter became principal, and he inspired the Church Missionary Society to extend the scope of the College to include the liberal arts as well as classics and theology. Suggestions that Fourah Bay College should be affiliated to an English university were made, and in 1876 affiliation to the University of Durham was established, whereby students could be admitted to any degree, diploma or licence of that University. At this time Sunter introduced extra-mural classes and some science teaching with the aid of Colonial surgeons who became part-time lecturers. Mathematics, and a little later geography, were subjects for degree courses.

In those days optimism in Sierra Leone was abundant, and the country was talked of as the place where West African development would start. There was even talk of a West African university developing from the College. The graduates of Fourah Bay College, with the standing of a British university degree, went out into the West African world and prospered, filling important posts in government, law, teaching, medicine, and the church.

However, the missionary finances started to dwindle, the Government did not recognize a great responsibility towards education, and the municipality of Freetown did not then feel a civic urge to develop or even maintain the College. Consequently the College was hard put to maintain itself; but it persevered with an average of twenty students until the end of the Second World War.

The first 'science' lecturer was appointed in 1924 and the foundation stone of a small block for science teaching was laid in 1925. From that time the establishment of the College has included a 'science' lecturer. This development was inspired and made possible by the bequests of two Africans: Dr. Obadiah Johnson and Dr. John Randall. It would seem that from 1929 the science lecturer was occupied with a few lectures given to the arts students, and with the non-graduate teacher training course which started in that year. In 1939 a course leading to the Durham Higher School Certificate was started with the hope that this would develop into a Faculty of Science. In 1941 the first students passed in botany, chemistry, physics and zoology—the teaching in all these four subjects being done by one lecturer.

On the outbreak of the Second World War the College buildings were requisitioned by the military authorities, and teaching continued in huts. In 1942 the College moved to Mabang in the Sierra Leone Protectorate some forty miles from Freetown. Here conditions were very primitive, science work proved impossible, and the science lecturer transferred to Government service and started the first formal sixth-form course in science at the Government Secondary School, Freetown. This lecturer was Dr. E. Downing, who eventually left Sierra Leone in 1946, but who returned in 1954 as head of the Department of Chemistry and dean of the Faculty of Science.

In 1945 a Commission to investigate higher education in West Africa, under the chairmanship of Sir Walter Elliott, visited the British West African Colonies, and as a result of this report, University Colleges and Colleges of Arts, Science and Technology were set up in Nigeria and the Gold Coast. When the Commission visited Mabang, economic conditions there were so difficult and the number of staff and students so reduced that they were not impressed and felt that the College was an uneconomic proposition and should be disbanded. This view was vigorously fought by two members of the Commission, Dr. E. H. Taylor-Cummings of Freetown and Sir James Duff, warden of the Durham Colleges. As a result of lengthy negotiations, a new ordinance was published in 1950 in which a largely autonomous College was set up, governed by a Council representative of Government, the Missions, the College teaching staff and other local interests. Financial responsibility was taken over by the Sierra Leone Government, which had the advice of the Council for Overseas Colleges of Arts, Science and Technology. The courses in the College were restricted to those already

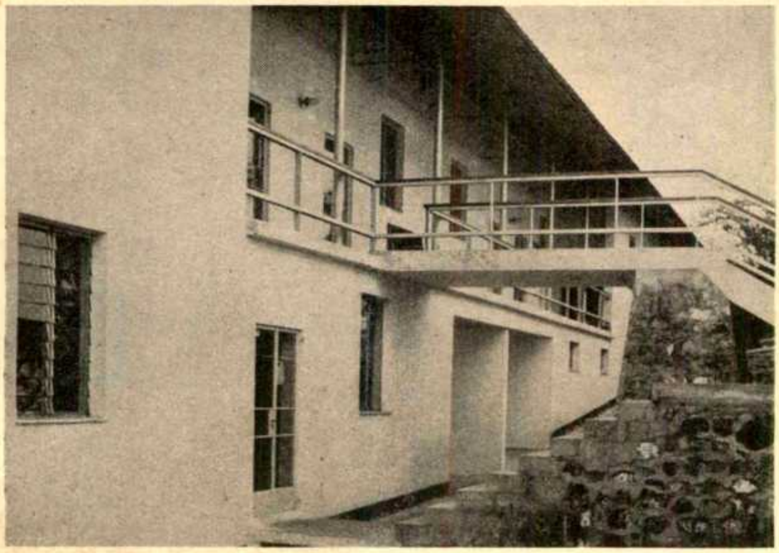

Fig. 2. The new building housing the Department of Zoology and part of the Department of Botany (Photo: T. Lloyd)

existing, namely theology, arts and economics as degree courses, a postgraduate diploma in education, a nongraduate teacher-training course, and science courses to General Certificate of Education (advanced level).

An Education Commission under the chairmanship of Mr. John Fulton (then vice-chancellor of the University of Wales) visited Sierra Leone in 1954 to inquire into the general educational needs of the country and to make recommendations for a long-term policy for the development of Fourah Bay College. In spite of restrictions, the College had grown in numbers. As a result of this the Commission suggested an expansion in the fields of pure and applied science and the goal of university status as an ultimate objective.

A further Commission under Dr. C. H. Wilson, vice-chancellor of the University of Leicester, reported in 1959, and in accordance with the recommendations of this Commission a request was submitted that the College be granted a Royal Charter incorporating it as the University College of Sierra Leone, and the Inter-University Council took over from the Council for Overseas Colleges of Arts, Science and Technology. At the same time the College's responsibility for non-graduate teacher training was transferred to a new and separate Teacher Training College. Now the College consists of Faculties of Arts, Economics, Theology, Pure Science and Applied Science, as well as a postgraduate Education Department and an Extra-Mural Department.

In 1946 the College had returned to Freetown from Mabang and exchanged its old site (where the Queen Elizabeth II Quay was built) for an old military hospital on Mount Aureol, a considerable area of plateau and hillside. The accommodation on this site was either in old stone buildings or temporary wooden structures. With the aid of grants from the Colonial Development and Welfare Fund, the Sierra Leone Government, and local firms, especially the Sierra Leone Development Co., new buildings designed by Mr. Frank Rutter are slowly replacing the old. One of the first priorities in the rebuilding has been new buildings for the science departments, and most of these are now completed (Fig. 2).

In 1955 there was one lecturer in each of the subjects of biology, chemistry and physics, and a teacher of general science, who ran a preliminary course with which to feed the science course. The Faculty of Science was then established containing the Departments of Botany, Chemistry, Geography, Mathematics, Physics and Zoology. Now each of these has a staff of three with the exceptions of Geography (two) and Mathematics (five). At the same time a Department of Applied Science was established, where students take a diploma in engineering, which was established after consultations with the Departments of Engineering, King's College, Newcastle. More recently the preliminary course in general science was discontinued, as the number of students who have taken sixth form science has increased.

Science teaching is concentrated now on the general B.Sc. degree of the University of Durham. The immediate developments envisaged are the consolidation of the present courses and departments, and the addition of honours degrees.

At the moment there are some 350 students registered in the College. As the College is fully residential, the number of students is at present regulated by hostel accommodation, though this is being increased in the near future. Students from Sierra Leone are given preference in acceptance, provided they are suitably qualified. At the moment about half the student body is Sierra Leonian, with a few students from Ghana and Gambia, and the rest from Nigeria. This large Nigerian population is in part a reflexion of the traditional association of that country with Fourah Bay College when the College was the only institution providing a university education in West Africa.

In the science departments a number of research projects are under way, most of them dealing with local conditions.

The biological departments have a wealth of material literally at their doorsteps—a forest reserve only 50 yd. from the Botany Department is being established. Research in the Botany Department under Dr. F. J. Taylor is being directed towards the physiology of the local flora and of freshwater algae, experimental taxonomy of the local flora, the ecology of soil fungi, and the ecology and microbiology of mangrove swamps. The head of the Zoology Department, Mr. J. Phipps, has been studying the local grasshoppers intensively, and other research in that Department includes the study of mud wasps with their spider food, and the nitrogen metabolism of stick insects. The Botany and Zoology Departments have recently established a Marine Biology Laboratory on the coast some eight miles from the College.

The Chemistry Department, under Dr. E. Downing, is interested in the chemistry of natural products, immediately in waxes and their component acid and alcohols. Future lines of development in this Department are likely to be influenced by the parent University.

The two main lines of research being established in the Physics Department (head of the Department, Mr. E. G. Dunstan) are the study of the Earth's magnetic field, which has developed out of studies carried out in Nigeria during the International Geophysical Year, and the study of atmospheric electricity, for which the tropics are especially suited.

In the Geography Department, under Mr. G. Dalton, the main topics of study at present are climatology and land-use surveys. A somewhat related topic under study in the Department of Applied Science, with Mr. B. Pesood, is the nature of lateritic soils, especially with regard to the application of cement stabilization techniques.

The conferment of university college status leads to the hope that Fourah Bay College will attract research workers in science wishing to work in the tropics. From the point of view of the biological field worker, for example, Freetown offers a unique annual rainfall regime on the coast of West Africa, and a range of varied habitats within relatively easy reach.

OBITUARIES

Academician I. V. Kurchatov

The death of I. V. Kurchatov on February 7 is a severe loss to physics, particularly in the U.S.S.R. As director of the Atomic Energy Institute of the Academy of Sciences and an outstanding leader in atomic energy research of all varieties, he played a leading part in determining Russian atomic energy policy. Much credit must go to him for the rapid post-war developments of the Russian programme, and he received many honours from the Soviet Government for his work. He was a member of the Praesidium of the Academy of Sciences and a deputy to the Supreme Soviet of the U.S.S.R.

Born in 1903 in Eastern Russia, Igor Vasilevitch Kurchatov graduated at the University of the Crimea in 1923 and started research at the Leningrad Physico-Technical Institute, then under the direction of A. Joffe, in 1925. Up to 1933 he carried out experimental and theoretical studies of dielectrics, particularly on the properties of Rochelle salt. He was the first to give a mathematical theory of ferro-electricity, basing his approach on the Langevin–Weiss theory of ferromagnetism. His many researches on this subject were summarized in a short book published in 1933. Other work at this time included studies of dielectric breakdown.

Kurchatov's nuclear physics work began in 1933, and in 1938 he was made director of nuclear physics research at the Leningrad Institute, where he and his collaborators conducted many pioneer researches into nuclear reactions produced by neutrons. His work on the activities induced in bromine, together with the results obtained by Fermi's group, led to the first identification of a long-lived nuclear isomer. Other work included detailed studies of the $^{10}B(n,\alpha)$ reaction, and the scattering and absorption of neutrons in water, paraffin and other moderating media. His group was thus exceptionally well placed to follow up the discovery of nuclear fission in 1939, and in 1941 he published an authoritative paper surveying various possible ways of developing a chain-reacting system based on thermal neutron fission of heavy elements.

The Atomic Energy Institute was started towards the end of the Second World War. It is a large establishment, occupying several hundred acres on the outskirts of Moscow, devoted to fundamental research on atomic energy. The wide scope of research there was revealed at the first Geneva Conference on the Peaceful Uses of Atomic Energy in 1955. From his public statement in 1958 claiming Russian priority in the development of a practical military form of the hydrogen bomb, it appears that Kurchatov played a leading part in military as well as civil applications of atomic energy.

In recent years, the problems of plasma physics and controlled thermonuclear reactions formed a significant part of the work at Kurchatov's institute. Studies of magnetic confinement of plasma were vigorously pursued there by a group led by a pre-war Leningrad colleague, L. A. Artsimovitch. The first public account of this work was given by Kurchatov at Harwell in May 1956, in a lecture remarkable not only for its content but also for the bizarre circumstance that his audience was at that time keeping work in this field secret. In 1958 he gave the first account of further work in his institute on this subject, including the building of the enormous magnetic trap installation *Ogra*.

Kurchatov's career is largely synonymous with the development of atomic energy in the U.S.S.R., and although he died before many important aspects have come to fruition, the Russian achievements in this field, together with the size and vitality of his research institute, stand as an impressive testimony to his abilities. He believed in the value of international collaboration in science, and gave a most friendly reception to visitors from England to his institute.

His high scientific abilities, integrity and personal charm greatly impressed all who met him.

R. S. Pease
B. F. J. Schonland

Prof. H. G. Robinson, C.B.E.

Henry Goland Robinson, first professor of agriculture in the University of Nottingham, for many years principal of the Midland Agricultural College and later director of the University of Nottingham School of Agriculture, died on February 20 at his home in Wellington, Shropshire, aged sixty-three. A graduate of the University of Durham, he held appointments at Cockle Park and the then University College of Reading, before becoming senior lecturer in agriculture at the Midland Agricultural College, Sutton Bonington, in 1925. Ten years later he was appointed principal of the Midland Agricultural College, and led the developments which culminated in its union with the then University College of Nottingham as the Faculty of Agriculture and Horticulture. Robinson became the first professor of agriculture and dean of the Faculty.

Robinson will long be remembered for his work for the Midland Agricultural College and the University of Nottingham School of Agriculture, and for his services to agricultural education and farming in Britain, for at one time he was a member of thirty-nine committees, including the Nottinghamshire War Agricultural Executive Committee and several of its sub-committees. He lectured on aspects of agriculture throughout Britain and was a keen advocate of good husbandry. For his services to agricultural education he was appointed a Commander of the Order of the British Empire in 1955, and he was elected emeritus professor of agriculture in the University of Nottingham on his retirement, through illness, in 1954. J. D. IVINS

NEWS and VIEWS

First Award of the Vetlesen Prize: Prof. Maurice Ewing

DR. MAURICE EWING, director of the Lamont Geological Observatory, Columbia University, New York, has been awarded the Vetlesen Prize for outstanding achievement in the earth sciences. This is the first award of this Prize, which consists of a gold medal, 25,000 dollars and support of publication of the recipient's scientific papers. Dr. Ewing, professor of geology at Columbia, and a native of Lockney, Texas, is known internationally for his work in the Earth sciences, and among the previous honours he has received are the Day Medal of the Geological Society of America; the Agassiz Medal of the U.S. National Academy of Sciences; and the William Bowie Medal of the American Geophysical Union. Dr. Ewing, who has been a teacher of both physics and geology, considers himself primarily a physicist who applies fundamental science—physics, chemistry and mathematics—to the study of the Earth.

The Vetlesen Prize was established this year at Columbia University by the G. Unger Vetlesen Foundation for "achievement resulting in a clearer understanding of the earth, its history or its relation to the universe". Like the Nobel Prizes established more than fifty years ago, the Vetlesen Prize recognizes no national boundaries, and any person in any country may be nominated for it. The Nobel Prizes, set up when the Earth sciences were of secondary importance, cover the fields of physics, chemistry, medicine, literature and peace. Funds for both prizes have been provided by Scandinavians. Alfred B. Nobel was a Swedish munitions maker. Georg Unger Vetlesen, a shipping executive, was a native of Norway who became a United States citizen during the Second World War. He established the foundation bearing his name shortly before his death in 1955. The Vetlesen Prize is to be awarded every two years provided a worthy candidate is presented.

The British Museum (Natural History): Sir Gavin de Beer, F.R.S.

THE retirement of Sir Gavin de Beer from the directorship of the British Museum (Natural History) at the end of April this year ends one of the most memorable decades in the history of this institution. He took office at a time when the Museum was still suffering from the effects of the War and during his tenure has succeeded in carrying out an ambitious programme of reconstruction and extension of the Museum facilities and also helped to restore its international prestige. The building of the North Block, with Lecture Theatre, Map Room, General Library and Reading Room, is one of the more obvious additions; but the Mammal Building, extension of the Whale Hall, completion of the Entomological Block, and the new Botanical Gallery, with Cryptogamic Herbarium and laboratories, have all greatly increased the amenities and facilities for scientific study. The Mammal Balconies now include exhibits on the functional morphology of mammals, and the Central Hall has been re-organized to accommodate the big pachyderms and a completely new set of exhibits on evolution which were specially prepared in connexion with the Darwin-Wallace Centenary.

Sir Gavin has carried out investigations on *Archaeopteryx*, and the evolution of Ratites, apart from many historical studies of special interest to the Museum, including the Darwin manuscripts and note-books; Sir Hans Sloane and the British Museum; the Banks correspondence; and he initiated the Historical Series in the Museum Bulletin. Among his other publications a "Short Guide to the Museum", "Scientific Research in the Museum", and a "Handbook to Evolution" were issued for the use of visitors. It was a fortunate coincidence that the Darwin–Wallace Centenary should have occurred during the term of office of one of the greatest authorities on evolution and he took a leading part in the celebrations of that event. In addition, Sir Gavin was president of the fifteenth International Congress of Zoology, held in London during 1958, which attracted by far the largest attendance on record. He was also president of Section D (Zoology) at the meeting of the British Association in Oxford in 1954.

Sir Gavin de Beer's successor will be Dr. T. C. S. Morrison-Scott, director of the Science Museum. Dr. Morrison-Scott was formerly on the staff of the British Museum (Natural History), where he worked mainly in the Mammal Department (see *Nature*, 177, 1061; 1956).

Laboratory of the Government Chemist: Mr. E. H. Nurse, C.B.E.

ON March 31, Mr. E. H. Nurse will retire as acting Government Chemist from what was until recently the Department of the Government Chemist. Mr. Nurse has spent the whole of his career in this Department, which he joined in 1915, following graduation with first-class honours in chemistry from the University of London. During the First World War he served for two years with the Royal Naval Air Service and the Royal Air Force as a hydrogen officer with airships. On the re-organization of the Scientific Civil Service in 1946 he became a principal scientific officer, and he was promoted to the grade of senior principal scientific officer in 1948. He has represented the Department on a number of scientific and technical committees and has been particularly concerned, both at headquarters and the subsidiary laboratories maintained at certain ports, with chemical work in connexion with revenue duties of Customs and Excise. He was appointed Deputy Government Chemist in 1954 on the retirement of Dr. J. R. Nicholls, and on the death of Dr. G. M. Bennett, in February 1959, he became acting Government Chemist. During his period as head of the

Laboratory he has seen its transfer from the Treasury to the Department of Scientific and Industrial Research. He was created C.B.E. in 1958.

Dr. D. T. Lewis

Dr. D. T. Lewis, who has been appointed Government Chemist, was born in Breconshire in 1909 and graduated at the University College of Aberystwyth with first-class honours in chemistry in 1930. He afterwards gained the degrees of Ph.D., and D.Sc., of the University of Wales. Prior to the War, he occupied various teaching posts and held lectureships at the University Colleges of Aberystwyth and Cardiff. In 1941, Dr. Lewis joined the Armaments Research Establishment of the Ministry of Supply and was associated with research and development work on conventional armaments. Leaving this work he was appointed one of the early members of Sir William Penney's atomic weapons team, and for the past seven years he has held the post of senior superintendent of the Chemistry Division at the Atomic Weapons Research Establishment, Aldermaston. In addition to his duties at Aldermaston he has also taken part in overseas trials in Australia and at Christmas Island. He has published a large number of papers in the various scientific journals dealing with inorganic physical and analytical chemistry. He has also published "Ultimate Particles of Matter", a book designed as an introduction for laymen to nuclear and associated sciences. Dr. Lewis is chairman of the Thames Valley Section of the Royal Institute of Chemistry and has been a representative of the Atomic Energy Authority on the board of governors of the Newbury Technical College.

Lucas Heights Research Establishment, Australia:
Dr. G. C. J. Dalton

Dr. George Clifford James Dalton has been appointed director of the Lucas Heights Research Establishment of the Australian Atomic Energy Commission. Dr. Dalton was born at Te Awamutu, New Zealand, in 1916 and was a Rhodes Scholar in 1936. From Oxford he went to the Royal Air Force where he was occupied with radar research. At the end of the Second World War he went to Harwell, where he was in charge of the Fast Reactor Group of the United Kingdom Atomic Energy Authority. This Group carried out the early scientific work which led to the decision to construct the fast breeder reactor at Dounreay. During 1949–55, he was professor of mechanical engineering at Auckland University, New Zealand. Dr. Dalton joined the staff of the Australian Atomic Energy Commission in 1955 as chief engineer and deputy chief scientist.

Second Nuclear Reactor for Lucas Heights

A second nuclear reactor is being installed at Lucas Heights by the Australian Atomic Energy Commission. In announcing this, the Minister for National Development, Senator Spooner, explained that it will be a small and simple apparatus by comparison with the Commission's first reactor, *Hifar* (see *Nature*, 181, 1433; 1958). It will be used in the Commission's general programme for the development of a high-temperature gas-cooled reactor and will be complementary to *Hifar*, which was a 10,000-kW. materials testing reactor used also for making radioisotopes. Production of isotopes, which has already begun, forms an important part of the Lucas Heights programme. The new reactor will produce 10 kW. and will be employed for many tests and measurements where the intense radiation of *Hifar* is unnecessary. These will include physics studies of reactor material, and sample cores (fuel assemblies) for future power-reactor systems. Because of its simplicity and inherent safety of operation, the new reactor will also be used for training staff who will eventually work on other reactors. It is expected that the cost of installing the new reactor and its associated facilities will be in the region of £65,000.

The Game Research Association

A national organization to be known as the Game Research Association has been formed, by a responsible group of farmers, landowners and others interested in game conservation, to promote game research in Britain. The Association will be financed entirely by donations and membership subscriptions, and will carry out research into all factors controlling the game population of the country. The subjects to be studied, as funds and staff become available, include ecology of partridges and pheasants in relation to agriculture, forestry and other land use, regarding game as a natural resource; and also the more immediate problems of game mortality such as gapeworm, the effects of toxic chemicals and effective re-stocking methods. Laboratories for a temporary research headquarters are being loaned by Imperial Chemical Industries, Ltd., at Fordingbridge. The president of the Association is Mr. Harold Macmillan, and vice-presidents, Sir John Ruggles-Brise and Major John Morrison; chairman of council, Lord Porchester. Mr. A. D. Middleton, director of the Imperial Chemical Industries Game Research Station, Fordingbridge, is honorary research adviser, and Mr. F. O. G. Lloyd is honorary secretary–treasurer (temporary London office at 7–8 Great Winchester Street, London, E.C.2).

The I.C.I. Game Research Station, Fordingbridge

The work of this Station at Fordingbridge, in Hampshire, will continue, but with some reduction in the research projects and more emphasis on advisory and educational work. Research will include methods of artificial propagation and population dynamics. The advisory staff will continue, through personal visits, literature and other means, to give practical application to the knowledge which has been gained during fourteen years of work at Fordingbridge and, in pre-war years, in Hertfordshire and other places. Naturally, Imperial Chemical Industries, Ltd., welcomes the formation of an independent Game Research Association, which should be able to expand research in this field. The Fordingbridge team will collaborate in every possible way in the work of the Association.

Research on Grouse

In addition to the Game Research Association, and the work of the Imperial Chemical Industries Game Research Station, a third research project, concerning grouse, is now firmly established. The ecological study of grouse in Scotland, under the direction of Prof. V. C. Wynne-Edwards, of the University of Aberdeen, financed for the past three years by the Scottish Landowners Federation, is now being taken over and expanded by the Nature Conservancy. This is to be part of a wider project known as the Unit of Grouse and Moorland Ecology, the first of such units to be operated by the Conservancy. Its research results, from a long-term study of grouse population dynamics, will inevitably be of permanent value to grouse management, which is an important feature of moorland economics.

State Aid for Museums and Art Galleries of Britain

FOR many years the Museums Association, through a succession of presidents and other officials, has emphasized the desirability—indeed the absolute necessity—of giving some measure of financial assistance to museums and art galleries, especially to the smaller provincial institutions. On February 26, Sir Edward Boyle, Financial Secretary to the Treasury, made a statement in the House of Commons which indicates that H.M. Government is at last fully alive to the present needs of museums and art galleries. There was a practical result in that various grants were announced. First was the direct provision in the Estimates for the national collections, which is to show an increase of £350,000 or 13 per cent. This welcome news will allow the British Museum to appoint fifty extra staff and enable the authorities to keep the Reading Room open to a later hour on, say, two evenings a week. Until 1959 the Victoria and Albert Museum, by means of its Circulation Department, was enabled through a Treasury Grant to disperse the hopelessly inadequate amount of £2,000 per annum to local museums for the purchase of specimens. Last year this sum was increased to £15,000, and now it is announced that the amount has been raised to £25,000. From the professional point of view, it was especially welcome news that the Minister of Education has decided to ask Parliament to approve of an entirely new type of grant to the Victoria and Albert Museum for the benefit of a provincial gallery. By this means it will be possible for Liverpool to purchase the "Holy Family" by Rubens.

It has been continually urged by the Government that the museum problem should be settled locally, and to this end the South Western Museum Regional Council was formed in January 1959. Its aim was to promote closer co-operation between museums, to improve the technical facilities available and to distribute financial assistance to museums and art galleries within the region. It was hoped that the Financial Secretary would be able to make a definite statement regarding assistance to this regional scheme, especially in view of the strong support given only the day before in a memorandum by the Association of Municipal Corporations. Unfortunately he was not able to announce any grant in this connexion. But perhaps the most welcome result of the debate in the House was the statement that the Government now realizes that many museums and art galleries in Britain are inadequately provided for, and that a general inquiry would serve a useful purpose. To this end the Chancellor of the Exchequer has invited the Standing Commission on Museums and Art Galleries to consult the Museums Association and others to consider the desirability of a complete review of provincial museums and art galleries. It is to be hoped that this review will take place in the foreseeable future.

Employment of Scientists in the United States

A REPORT issued by the National Science Foundation gives final information on about 116,000 employed scientists (out of about 127,000) in the National Register of Scientific and Technical Personnel for 1954–55, roughly half of all American scientists at the time (American Science Manpower: Employment and Other Characteristics, 1954–55. Pp. vi+48. (N.S.F. 59-39.) Washington, D.C.: National Science Foundation, 1959). The largest group (41,000) was employed in chemistry and chemical engineering. About half of the 116,000 were employed in private industry and nearly one third by educational institutions. About 50,000 (44 per cent) were engaged in research, development or field exploration, about 20,000 in management or administration and about the same number in teaching. Nearly 40 per cent of the scientists held a doctorate and more than 33 per cent a master's degree. In 1954–55 the median annual salary for scientists reporting this item was 6,525 dollars and, in general, salaries were highest for scientists with a doctorate and those employed in private industry. About 8,000 (about 7 per cent) of the scientists were women, who were employed chiefly in the biological sciences, chemistry and psychology. Of the 127,000 respondents in the 1954–55 Register, more than 90 per cent were civilian scientists employed in scientific or in non-scientific specialities, including education, social work, and humanities. Of the remainder, about 4,000 were on active military duty, 4,200 students, 700 unemployed and 2,000 retired. These groups are not covered in the report.

U.K. Government Relief for Scientific Societies

THE annual report for 1959 of the Parliamentary and Scientific Committee, besides a concise summary of the addresses and discussions during the year, records that the Committee's Rating Sub-Committee welcomed the recommendations of the Pritchard Committee insofar as they offer a 50 per cent rate relief to scientific societies and similar bodies, and extends to industrial research associations the same degree of rating relief as applies to industry in general (pp. 28. London: Parliamentary and Scientific Committee, 1960). Subject to certain reservations, the Committee hopes that the necessary legislation will be introduced by the Government at the earliest possible opportunity. First, the Committee regrets that the Pritchard Committee's recommendations, if implemented, will impose a material rate burden on those scientific organizations which have obtained total exemption under the Scientific Societies Act, 1843, and will also debar from such full relief other scientific societies which might have been able to qualify for exemption under that Act at some future date. The Committee recommended accordingly that if the Government did not find it possible to enable scientific societies to continue to enjoy full rating relief, arrangements should be made by the Minister for Science to afford assistance to scientific societies which will be rendered less effective by the implementation of the Pritchard Committee recommendations. Secondly, the Committee submits that certain modifications in the precise recommendations of the Pritchard Committee will be necessary to ensure that some scientific societies are not debarred from the rating relief recommended by reason of the definition of what is to constitute a charity for the purpose of rating benefit.

Vegetable Research

THE Research Station at Wellesbourne, with its sub-station at Paglesham, Essex, was established in 1949 and has now a considerable volume of results (Ninth Annual Report, 1958, of the National Vegetable Research Station, Wellesbourne, Warwick. Pp. 55. 1959). A long-term assessment of chemical, physical, biological and ecological changes involved in raising soil fertility to the high level required for vegetable production shows that farmyard manure exerts the greatest single effect. For some crops, however, the greatest yield is only obtained by supplementing with fertilizer. More detailed chemical investigations showed that there was an accumulation of sucrose, fructose and glucose, and a decrease in phosphorylated intermediaries, in the roots of beet immediately before flowering.

The report describes many studies on competition between weeds and crop, and the effects of pre-planting treatments. Various agents for weed control are under investigation, but it is interesting to note that the mere fact of growing vegetables for three years has reduced the number of weed seeds in the soil to about one-sixth the original number. Studies on the control of environmental conditions include extensive work on irrigation, and lettuce particularly benefited from adequate watering through its period of growth. Disease studies are now extensive, but perhaps the most significant findings are that silvering disease of beet can be controlled by seed treatment with streptomycin, the effectiveness of aldrin in reducing club-root disease, and the use of zinc frit for the control of powdery mildew of turnips and cabbage. Entomological investigations are concerned mainly with cabbage root fly, carrot fly and the root aphid of lettuce, while in methods of control, seed treatment and soil incorporation figure largely. Plant breeding is perhaps slow to give immediate results, but the report shows useful progress with many vegetables ; in this, as indeed in other sections, reference must be made to the report itself, and it is clear that as the years go by it will become more and more valuable.

Plasma Physics and Thermonuclear Research

The new section, Part *C*, of the *Journal of Nuclear Energy* is to be published quarterly and is devoted to plasma physics, accelerators and thermonuclear research (Pergamon Press, 4–5 Fitzroy Square, London, W.1). As Sir George Thomson explains in his brief foreword to the first issue (1, Nos. 1/2, October 1959), plasma physics is part of that study of gaseous discharges to which physics owes the fundamental discoveries of electrons and of isotopes, and from which it is hoped to be able to solve the problem of obtaining controlled energy from fusion reactions between nuclei of isotopes of hydrogen. Prof. M. S. Livingston in his foreword explains that accelerator technology has contributed significantly to the newer technology of thermonuclear science, and therefore the new journal will perform an important and useful function in sponsoring the merger of the two technologies with the study of plasma physics. In addition to eight original contributions, the October issue contains abstracts in English, prepared by the U.S. Atomic Energy Commission, of articles from "The Physics of Plasmas", published by the U.S.S.R. Academy of Sciences, a collection divided into four volumes in which are described the theoretical and experimental investigations carried out during 1951–58 in the Institute of Atomic Energy of the U.S.S.R. Academy on controlled thermonuclear reaction and associated subjects. Other features of the issue are translations of articles from *Atomnaya Energiya* and a list of titles of current papers and reports on plasma physics, accelerators and thermonuclear research appearing in some 50–100 journals published throughout the world. The October list covers publications since January 1, 1959. The editor-in-chief of Part *C* is Dr. J. V. Dunworth.

Russian Chemical Journals

The Chemical Society announces that cover-to-cover translations of two additional Russian chemical journals are to be published with the assistance of the Department of Scientific and Industrial Research. The *Russian Journal of Physical Chemistry* (*Zhurnal Fizicheskoi Khimii*) is being published by the Society in translation starting with the issue for July 1959. It is intended that subsequent monthly issues will appear regularly, and it is hoped that the interval between the receipt of the Russian original and the publication of the translation will eventually be reduced to about three months. Mr. R. P. Bell is scientific editor of the translation. Another journal, *Uspekhi Khimii*, will be published monthly in translation under the title *Russian Chemical Reviews*, starting with the issue for January 1960, which it is hoped to make available in April. The scientific editor of the translation will be Dr. J. N. Agar, assisted by Dr. A. R. Katritzky and Dr. R. H. Prince. Translations for these journals are supplied for the Society by Infosearch, Ltd., and distribution is undertaken by the Cleaver-Hume Press, Ltd. Subscription inquiries should be addressed to the sole distributors, Cleaver-Hume Press, Ltd., 31 Wright's Lane, London, W.8.

The United States National Museum

In a comprehensive report for 1959 it is stated that the site near the Washington Monument is now being prepared for the proposed Museum of History and Technology (The United States National Museum. Annual Report for the year ended June 30, 1959. Pp. vi+178. Washington, D.C.: Government Printing Office, 1959). The policy of this Museum is described as "to illustrate by means of the historic collections in the Smithsonian the cultural and technological development of the Nation since Colonial times". The plans include some thirty-eight major halls which have been coherently related to show a logical sequence of events. The needs of the individual museum visitor have been kept in mind, for it is conservatively estimated that some five million persons will enter the building each year. Provision is made for adequate air conditioning for the protection of objects as well as the comfort of visitors, circuits for television and electronic guide devices, and other electrical and plumbing devices in this great modern display unit. The plans thus indicate a museum building that will for the first time allow the great collections of the Smithsonian to be viewed in an adequate manner. During the sixth year of the continuing modernization programme work has also moved steadily forward on the other two fronts, namely, the renovation of the permanent exhibition halls in existing buildings and assistance to other branches of the Smithsonian Institution in the planning and presentation of exhibits.

Proteins from Grass

Pirie and other scientific workers have directed attention to reserves of protein available in grass and other chlorophyll-containing material normally consumed as animal foodstuffs, but potentially of use to man. The efficient production of good-quality edible protein from such sources depends on the solution of various biochemical engineering problems which have been investigated by the Rothamsted group for some years. A recent report from British Glues and Chemicals, Ltd., reflects the increasing interest in leaf protein; the Chayen impulse process, introduced a few years ago by this firm for the disintegration and extraction of bones and animal fats, has now been applied to a variety of other materials, including oilseeds, such as groundnuts, and grasses. The principle of this method lies in the use of shock waves of sufficient intensity and frequency to burst the tissues and cells of biological materials; it is illustrated by the depth-charge technique used in anti-submarine warfare and by the method of fishing with the aid of a dynamite charge. The report states that 100 tons of fresh grass (of 80 per cent water content) treated by the Chayen method yields about 2¼ tons of edible protein together with two important by-products—7 tons of fibre (suitable for the production of hardboard) and 0·6 tons of a 'chlorophyll syrup' containing fat-soluble vitamins, pigments and steroids. The commercial development of this process will be watched with interest by all who are concerned in any way with the nutritional problems of tropical and other areas, and it is to be hoped that the method can be introduced into those regions where additional supplies of supplementary protein are most needed.

Department of Scientific and Industrial Research: Studentships and Fellowships

THE Department of Scientific and Industrial Research, which this year has awarded studentships to 2,143 research workers, as well as 271 advanced course studentships and 48 research fellowships, at a cost expected to exceed £1 million, and grants totalling £2,238,000 to 287 investigators at universities, technical colleges and similar institutions for researches in the natural and human sciences and technology, has now issued two publications replacing the former "Notes on D.S.I.R. Grants to Graduate Students and Research Workers". "D.S.I.R. Studentships and Fellowships" describes in detail the different types of studentships and fellowships in science and technology offered by the Department, including those tenable abroad and also the science studentships and fellowships administered on behalf of the North Atlantic Council (pp. 24. 1*s*. 6*d*. net. D.S.I.R. Research Grants. Pp. iv+10. 1*s*. 3*d*. net. London: H.M. Stationery Office, 1960). "D.S.I.R. Research Grants" gives similar information about the grants for special researches awarded by the Department and includes a note on the scheme of Senior Visiting Fellowships instituted by the Organization for European Economic Co-operation which the Department operates.

Gassiot Research Fellowships in the Meteorological Office

THE official list of the functions of the Meteorological Office includes "Research in Meteorology and Geophysics". In the long history of the Office, 'geophysics' has come to mean terrestrial magnetism and seismology, and continuous observations in these fields have been made for many years at Kew, Eskdalemuir and Lerwick. In the past two decades the demands of meteorology have been such that it has not been possible to do much more than to maintain and publish the observations, especially in seismology. It has long been recognized that this is not a desirable state of affairs, and an important step towards the revival of research in the Meteorological Office in both fields has now been taken by the creation by the Secretary of State for Air of two research fellowships (named after John P. Gassiot, F.R.S., who in 1871 bequeathed to the Royal Society a large sum of money for the maintenance of magnetical and meteorological observations at Kew) in geomagnetism and seismology, respectively. The fellowships, which are tenable for seven years with a possible extension to ten years, will allow the holders to pursue any studies falling within the acknowledged fields of the two disciplines, with the aid of facilities provided by the Meteorological Office. It is also intended that there will be close contact with universities. Selection of the Fellows will be made in association with the Royal Society.

Gift to the Rensselaer Polytechnic Institute

A GIFT of one million dollars has been pledged to the Rensselaer Polytechnic Institute by Mr. and Mrs. Erik Jonsson, of Dallas, Texas, through the Jonsson Foundation, to help finance a new Science Centre at Rensselaer. Mr. Jonsson is chairman of the board of Texas Instruments, Inc. The gift, one of the largest single grants recently received by the Institute, is for construction of one unit of the Science Centre on the Rensselaer campus.

University News: Birmingham

UNDER the policy of general expansion throughout the University of Birmingham, it is now announced that the Faculty of Commerce and Social Science will be accommodated in a completely new building, and plans are in preparation with the view of commencing building work in 1961 and completing it in July 1962. Work on the £1 million building for the Department of Biology will start in mid-1960 and will be fully completed by 1963.

Dr. J. T. Davies, lecturer in chemical engineering in the University of Cambridge, has been appointed to the chair of chemical engineering and to the headship of the Department of Chemical Engineering from October 1, 1960. The following appointments have also been made: Dr. Gillian Vaisey, to be lecturer in the Department of Civil Engineering; G. Lees, to be lecturer in road materials in the Department of Civil Engineering; D. R. Milner, to be Lucas Fellow in metal joining in the Department of Industrial Metallurgy. A readership in reactor physics has been conferred on Dr. J. Walker, senior lecturer in physics, and in social medicine on Dr. C. R. Lowe, senior lecturer in social medicine.

Leeds

APPOINTMENTS to professorships are announced as follows: Dr. J. Nutting, as professor of metallurgy from a date to be arranged; Mr. D. R. Wood, as professor of pharmacology from September 1; and Mr. J. M. Cameron, as professor of philosophy. Major grants to the University are: by the Department of Scientific and Industrial Research, £40,000 to the Department of Physics for research on cosmic ray showers, £5,650 to the Department of Leather Industries, and £2,200 to the Department of Electrical Engineering; by the Ministry of Aviation, £2,000 to the Houldsworth School of Applied Science for research on the fatigue properties of ferrous and non-ferrous alloys; by Imperial Chemical Industries, Ltd., £1,500 to the Department of Inorganic and Structural Chemistry to support X-ray structure analysis; and by the Hickson's Timber Impregnation Co. (G.B.), Ltd., £1,000 to provide a fellowship in the Department of Botany.

South Africa and Commonwealth Scholarships

MR. I. F. A. DE VILLIERS, First Secretary in the Office of the High Commissioner in London for the Union of South Africa, writes: "The article in *Nature*, of January 30, p. 271, on 'The Wandering Scholar in the British Commonwealth', mentions references, in debates in both Houses of Parliament, to the non-participation of the Union of South Africa in the Commonwealth Education scheme. I am concerned that this may have conveyed the incorrect impression that South Africa does not intend to participate; the actual position is that while the South African Government was not able to give consideration to the proposals of the Oxford Commonwealth Education Conference until quite recently, it has in fact decided to participate in both the Commonwealth Scholarship Plan and the Commonwealth Education Liaison Committee."

Announcements

PROF. LEO SZILARD, professor of biophysics at the University of Chicago, has been awarded the Albert Einstein Gold Medal and Award for 1960. The Award itself is valued at 5,000 dollars, and is presented annually with the Medal.

THE award of the Birbal Sahni Medal for 1959 has been made to Prof. P. Parija. The award was instituted by the Indian Botanical Society through a grant of Rs. 2,500 made by Prof. T. S. Sadasivan (Madras) in 1957 in memory of the late Prof. Birbal Sahni. The Medal is awarded annually in recognition of services to botany in India.

MR. R. M. WYNNE-EDWARDS, joint managing director of Constructors, John Brown, Ltd., has been appointed chairman of the Building Research and Road Research Boards for the period April 1, 1960–March 31, 1965, in succession to the two present chairmen, Sir Herbert Manzoni and Mr. E. John Powell, who have completed their terms of office.

THE editorial office of *The Biochemical Journal* has moved from the Lister Institute to 133–135 Oxford Street, London, W.1, where all further communications should be sent.

BY the generous benefaction of Prof. R. L. Wain, professor of agricultural chemistry in the University of London (Wye College), an income from the patents on his discoveries in the field of selective herbicides has been made available to the Agricultural Research Council and has been used to establish the Wain Trust Fund. One of the objects of the Fund is to facilitate the interchange of scientists between universities in the United Kingdom and overseas. The Council can now make available annually one to two awards (depending on the sums involved) to members of university staffs in Britain who wish to spend a period abroad. Applications or inquiries should be made to the Secretary, Agricultural Research Council, Cunard Building, Regent Street, London, S.W.1.

THE forty-fourth annual meeting of the Federation of American Societies for Experimental Biology will be held in Chicago during April 11–15. Further information can be obtained from Sidney S. Negus, Medical College of Virginia, Richmond, Virginia.

THE spring meeting of the Photobiology Group will be held at the University of Reading during April 1–2. Further information can be obtained from Dr. D. Vince, Department of Horticulture, University of Reading, at Shinfield Grange, Shinfield, Berks.

THE Department of Chemical Technology of the Bradford Institute of Technology is organizing a symposium on Fluorescence and Fluorescent Compounds on May 6. Further details can be obtained from the Registrar, Institute of Technology, Bradford, 7.

IT is announced that the third international congress in Photobiology is to be held in Copenhagen during July 31–August 5. The last date for registration is April 1, and further information can be obtained from Dr. B. C. Christiansen, Finsen Memorial Hospital, 49 Strand Boulevard, Denmark.

THE eighth conference of the British Agricultural History Society will be held during April 7–9 at Harper Adams Agricultural College, Newport (Salop). Further information can be obtained from the Secretary of the British Agricultural History Society, Department of Agriculture, University of Oxford.

THE Institution of Mining and Metallurgy will hold an international Mineral Processing Congress at Church House, Westminster, London, S.W.1, during April 6–9. Further information can be obtained from the secretary, B. W. Kerrigan, Institution of Mining and Metallurgy, 44 Portland Place, London, W.1.

THE Faraday Society has announced that a general discussion on Oxidation Reduction Reactions in Ionizing Solvents will be held in the University of Durham, King's College, Newcastle upon Tyne, 1, during April 11–13. Further information can be obtained from the Assistant Secretary, Faraday Society, 6 Gray's Inn Square, London, W.C.1.

THE second International Spring School of Physics of the Istituto di Fisica Theorica of the University of Naples will be held during April 20–May 7. The school is sponsored by the Società Italiana di Fisica and the University of Naples, and is to receive a grant-in-aid from Nato. Applications must be sent to the director of the School not later than March 31.

THE sixty-fifth annual congress of the South-Eastern Union of Scientific Societies will be held during April 22–25. Lectures and excursions will be held covering the biology, zoology, geology and archæology of the south-eastern counties; visits to various museums in the region will also be made. Further information can be obtained from F. Edwards, 53 The Drive, Shoreham-by-Sea, Sussex.

THE annual conference of the Ergonomics Research Society will be held in Cambridge during March 28–31. The topics to be discussed include ergonomics in the Armed Forces, in building research and athletics; and ergonomics in the Netherlands. There will be a symposium on factors in prolonged work, especially inspection. Further particulars can be obtained from Mr. D. U. Hunt, British Iron and Steel Research Association, Park Lane, London, W.1.

THE Annual Easter Congress of the Universities Federation for Animal Welfare will be held at Saltley College, Birmingham, during April 8–12. The programme includes a symposium on Humane Experimental Technique and lectures on introducing animal welfare to Libya (Dr. F. Jean Vinter), aspects of meat production (D. Norma Egginton) and humane slaughter (Colonel D. J. Anthony). Further information can be obtained from UFAW (Easter Congress), 7a Lamb's Conduit Passage, London, W.C.1.

DEVELOPMENT OF WOOD-USING INDUSTRIES

UNDOUBTEDLY among the most significant happenings of the past twelve months for the future of forestry and timber in Great Britain has been the publication of two reports on economic studies carried out by a Canadian firm of consulting engineers, Messrs. Sandwell and Co., Ltd., of Vancouver. These reports are entitled "Small Pulp Mill Survey" and "Board Mill Survey", respectively, the former having been conducted under the auspices of the Organization for European Economic Co-operation, the United Kingdom being one of seven member countries studied, while the latter was undertaken for selected regions in Great Britain at the request of the Forestry Commission[*].

There are two reasons why such surveys were called for. First, the wood-pulp industry in Europe is very largely concentrated in Scandinavia, the remaining countries importing either pulp or materials made from it, such as paper or fibreboard; naturally, it was desirable to see if something could be done in the home country to reduce dependence on imports and to develop new industries. Secondly, as a consequence of the extensive afforestation programmes of the past three or four decades, a very large volume of wood is becoming available, mostly from the small trees removed in the thinnings that must be carried out to make it possible for the trees retained in the forest to grow to saw-timber size within a reasonable time. This amount is not only already large, but also is increasing annually—and will continue to do so for a long time yet—while one of its main markets, namely, pit-wood for the coal mines, is approaching saturation; Scotland's needs are already met entirely from Scotland's own forests, and the demand is showing signs of falling off.

Basic to these studies are the facts that Britain imports three-quarters of a million tons of paper and boards out of her annual consumption of 4 million tons (1956 figures), and production in Britain of the balance is dependent on the import of 2 million tons of pulp. Expressed otherwise, three-quarters of the wood that is consumed by the British paper industry is imported.

It has always been recognized that for a pulp mill to be profitable there are several essential requirements which are not easy to fulfil. Only three of these requirements need be mentioned here: (1) a large and regular supply of suitable wood; (2) the cost of that supply delivered at the mill must be low compared with the price that is paid in the existing markets for such material for other uses such as pit-wood, light poles and box-wood; (3) a good water supply with effective means of disposal of effluents.

The consultants have tried to answer two main questions: (1) Given a delivered cost for wood, what is the minimum capacity of a pulp mill to be economically feasible under any of the processes available? (2) If the output is to be as low as 30 tons per diem, at what price must wood be available? It is not possible here to discuss the details; but their findings are that a separate plant set up solely to produce ground-wood (that is, mechanical) pulp is out of the question; that for a chemical (bleached sulphate) pulp plant receiving its raw material at a price that would be viewed as so low as to be doubtfully attainable by United Kingdom growers—though it is the average top price prevailing in North America—namely, £8 10*s*. a ton, the plant would have to be able to turn out at least 80 tons of pulp a day. If its capacity were as low as 30 tons a day, the price that it could afford to pay for the wood would be altogether impossibly low.

The overall conclusion is that if Britain is to develop in this field, her pulp mill must be integrated with other wood-based industries, notably with paper-making, like the recently opened hardwood pulp mill at Chepstow. Integration with saw-milling and connected industries such as veneering, plywood and furniture would also be helpful by providing an appreciable proportion of the raw material from waste available on the spot at a very low cost. If this can be done, a relatively small mill of 30–40 tons capacity should be able to pay a price for the rest of its wood which, though still lower than is currently being obtained, can be viewed as not impossibly so.

It is estimated that by 1965 two centres in Scotland (Inverness and Fort William) would probably be producing enough wood surplus within 100 miles radius by road to support a small-scale pulp mill if it can pay something like the current price for its wood, but probably only ten years later if its price is appreciably lower. At least two other centres are expected to have a similar surplus by 1975. It is known that a proposition to site such a mill in Scotland is under serious consideration by a leading group of wood industrialists. Incidentally, it has to be noted that our paper is mostly made of blended materials, including large amounts of straw—of which much more would be available if needed—and imported pulp of a type that would be selected in relation to the properties of that obtained from the local materials.

The second survey deals with the possible establishment of further hardboard, chipboard and insulating board mills, using the same raw material from the thinnings of our plantations. Although the amount of hardboard we import to-day corresponds to a production of 500 tons per diem, it is concluded that its manufacture would not be economically attractive unless the raw material could be cheapened to an extent that, as for the pulp mill, would only be attainable by integration with other wood industries, and this is not currently a practicable proposition with sawmilling because this industry is at present dispersed among a very large number of relatively small units. It is emphasized that chipboard and insulating board mills also require to be large to be reasonably profitable, with a daily output of at least 50 tons. They would be able to pay a rather higher price for their wood than that quoted for the pulp mill, a price that is still lower than the grower is at present getting, but within the range that he might be able to accept without putting himself out of business, provided every possible economy was made. Capital requirements would be about £1–2 million. In contrast with the prospects of further pulp mills, the most favourable area for board mills is considered to be southern England.

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[*] (1) Forestry Commission. Small Pulp Mill Survey, United Kingdom—Economic Study. By Sandwell and Co., Ltd. Pp. iv+48. 4*s*. net. (2) Board Mill Survey, United Kingdom—Economic Study. By Sandwell and Co., Ltd. Pp. vi+70. (London: H.M. Stationery Office, 1959.) 5*s*. net.

In the light of these surveys, it seems likely that Britain will have new wood-using mills of either or both types being built in the coming decade. There are not a few criticisms of the premises underlying these Sandwell reports with regard to costs, prices and trends, and suggestions that the size for profitability, at least for board mills, could be rather smaller and the capital cost less. However, it seems agreed that there must be a considerable reduction of the costs of growing and delivering the large amounts of wood required, and that there is certainly room for reduction by improved efficiency all along the line; there will be every incentive for cheaper production as other markets are bound to fall during what, without such mills, will be a period of large over-production. In all cases there will be marketing problems, especially in view of the free competition of imports under the Outer Seven trade agreements, but cautious optimism appears justifiable, and expansion in the directions indicated is to be expected in the near future.

H. Champion

BIOCHEMISTRY AT THE IMPERIAL COLLEGE OF SCIENCE AND TECHNOLOGY, LONDON

THE Imperial College of Science and Technology, London, has accepted a benefaction of £350,000 from the Isaac Wolfson Foundation to build and equip new laboratories for biochemistry and chemical microbiology and to support teaching and research in these subjects. Dr. E. B. Chain has been appointed to the chair of biochemistry and will take up the appointment in 1961.

This munificent benefaction will enable the College to erect a new building, to be known as the Wolfson Laboratory, adjacent to the Royal College of Science building in Imperial Institute Road. The Biochemistry Department will also occupy some laboratories in the Royal College of Science building shortly to be vacated by the Physics Department for its large new building in Prince Consort Road. Biochemistry at the College, which has been under the direction of a reader since the last occupant of the chair, Prof. A. C. Chibnall, succeeded Gowland Hopkins at Cambridge during the War, will thus burgeon out. The Department's new equipment will include fermentation units of comparatively large size and workshops for advanced mechanical and electronic equipment. It will provide accommodation for training and research in the neglected borderland between biochemistry and chemical engineering, and for research on an adequate scale on the biochemical production of chemicals of interest. Such a department, set in the Imperial College, with its very large school of engineering and flourishing schools of physics, organic chemistry and plant physiology, should find conditions highly propitious for major advances in biochemical knowledge.

Dr. Chain has since 1948 been at the Istituto Superiore di Sanità in Rome, where he has built up one of the best equipped, largest and most active centres of biochemical research in Europe. He has had there many of the facilities for which he had earlier, and unsuccessfully, pressed in Great Britain—in particular, equipment of pilot-plant scale for fundamental studies of substances of microbial origin and biochemical interest.

Dr. Chain, a graduate of the University of Berlin, was forced to leave Germany by Nazi persecution in 1933, and he became a British subject in 1939. He first worked under Gowland Hopkins at Cambridge, on snake venoms and phospholipids. In 1935 came an invitation from Prof. (now Sir Howard) Florey to join the scientific staff of the Sir William Dunn School of Pathology at Oxford, with the object of organizing a biochemistry section there. As University lecturer in chemical pathology he remained at Oxford until the end of 1948. His work there included a study of the biochemical mode of action of some snake venoms, on the mode of action of lysozyme, on the mode of action of the spreading factor, on tumour metabolism and on a rapid method for the determination of phosphate. In 1938, with Florey, a systematic study of anti-bacterial substances produced by micro-organisms was started. This led to the discovery of the curative properties of penicillin and heralded the era of antibiotics. In this work he played a leading part in the chemical studies on the isolation and structure of penicillin. For his part in the discovery of penicillin he was awarded the Nobel Prize for Medicine and Physiology in 1945, jointly with Sir Alexander Fleming and Florey. Dr. Chain was elected to fellowship of the Royal Society in 1949.

At the Istituto Superiore di Sanità, with the support of Prof. D. Marotta, director of the Institute, and the Italian Government, Dr. Chain has carried out research in many directions. His collaborators have included chemical, mechanical and electronics engineers, biochemists, organic chemists, microbiologists and geneticists. A wide range of subjects has been studied, mainly in two fields of biochemistry, chemical microbiology and intermediate carbohydrate metabolism in animals, touching on the methodological, biochemical and biological as well as the chemical engineering aspects of the subjects.

Among the biochemical subjects studied in the field of chemical microbiology have been factors involved in the biosynthesis of penicillin. The continuation of this work led to the discovery, by a team of research workers in the Beecham Laboratories in England, to which Dr. Chain has acted as adviser and consultant, that the main part of the penicillin molecule (6-aminopenicillanic acid) could be made by biosynthesis, and that this could be modified afterwards by chemical means. A great range of new antibiotics can be made in this way. They include a penicillin which is acid-stable and can be taken by mouth and one which attacks staphylococci resistant to normal penicillin. Compounds of this kind open up exciting prospects in the antibiotics field.

Other studies conducted in Rome have been on the green iron-containing streptomycete pigment ferriverdin, on submerged kojic acid and citric acid fermentations and on the production of ergot alkaloid in submerged fermentation. Among the chemical engineering aspects of chemical microbiology there have been extensive studies on agitation and aeration. Biological studies have included investigations on the relations between morphology and metabolism in filamentous fungi and genetical problems in filamentous fungi. Studies on carbohydrate metabolism in animals have been concerned with the mode of action of insulin and the interrelations between carbohydrate and amino-acid metabolism in brain and nervous tissues.

DIRECTION OF SCIENTIFIC AND TECHNICAL RESEARCH IN FRANCE

A PUBLICATION, *French Science News* (2, April–June, 1959), published by the Direction Générale des Affaires Culturelles et Techniques for the Ministère des Affaires Étrangères, describes the organization which has been set up to determine policy for scientific and technical research throughout the country. Two high-ranking committees have been formed, the first being an Interministerial Committee, the members of which include the Prime Minister as chairman, the Minister of National Education, the Minister of the Armies, the Minister of Finance and Economic Affairs, the Minister of Industry and Commerce, the Minister of Agriculture, the Minister of Public Health, and qualified representatives of the French community.

The main tasks of the Interministerial Committee, which is responsible for proposing to the Government all measures for the development of scientific and technical research, are as follows:

"To maintain a fair balance between the amount of credits appropriated for the different fields of research;

"To develop to its maximum usefulness French national research, which presupposes a detailed study of the profession of a research specialist and the milieu in which research is performed;

"To anticipate new research possibilities which will benefit the nation, which presupposes long-term government options in the field of science."

The second is an Advisory Committee, the members of which are: Maurice Letort as chairman (Letort is scientific director of Research and Studies, Centre of Charbonnages de France); Prof. Pierre Aigrain, vice-president of the Paris Faculty of Science; Prof. Jean Bernard, Paris Faculty of Medicine; Prof. Rene Dumont, National Agronomic Institute; Prof. Paul Germain, Paris Faculty of Science; Rene Latarjet, co-director of the Radium Institute and the Curie Foundation; Prof. Andre Lichnerowicz, Collège de France; Maurice Ponte, general director of the Compagnie Générale de T.S.F.; Prof. Charles Sadron, Strasbourg Faculty of Science and director of the Centre of Macromolecular Research; Pierre Taranger, industrial director of the French Atomic Energy Commission; and Felix Trombe, research director at the National Scientific Research Centre (Mont-Louis).

The Committees are served by a common secretariat directed by a general delegate. The general delegate can form work committees for specific objects and composed of experts. He works in close co-operation with the general planning commissioner.

The journal is published quarterly by the Association pour la Diffusion de la Pensée Francaise, 23, rue La Perouse, Paris, 16^{e}.

PRODUCTION AND CONSUMPTION OF ENERGY IN GREAT BRITAIN

DURING the past four years some remarkable changes have taken place in the consumption of energy in the United Kingdom. Indeed, since the War the situation in the whole of Western Europe has changed, from that of an energy-exporting economy to that of an energy-importing one. A few years ago there were fears of a shortage of energy in Great Britain. To-day there is the picture of rapidly rising coal stocks, and of real alarm in the mining villages.

Recently this subject was dealt with in an enlightening talk to the Fuel Luncheon Club by Sir Harold Hartley*, whose association with the international study of the energy problems of Western Europe entitle him to be listened to with earnest attention. His revelations should disturb any complacency with regard to the extent of our technological effort in the development of our coal resources.

The pattern of the energy consumption has changed quickly in the following respects in the past four years. Coal consumption fell 3 per cent per annum between 1955 and 1959, while that of oil rose by 12 per cent per annum. Electricity consumption continued its steady annual rise of 6 per cent, while gas declined by 1 per cent per annum. The competitive position depends upon factors of efficiency and of convenience, the relative basic prices of the therm in the primary forms of energy being: coal at the pithead, price 3½*d*., at £10 per ton, 8¼*d*.; fuel oil, at £10 per ton, 6*d*.; paraffin, at £20 per ton, 11*d*.; while gas at the average price costs 1*s*. 9*d*. per therm, and electricity at 1·55*d*. per unit, 3*s*. 9*d*.

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* Talk to the Fuel Luncheon Club on January 19 on "Energy in the United Kingdon—the Changing Scene", by Sir Harold Hartley.

While stagnant demand has struck the coal-fields, technical advancement has raised the potential productivity of the deep mines to a figure of 3·86 tons per man-shift. This is a pointer to the possibilities of saving the competitive position of coal so far as regards price. The technological aspects offer greater possibilities, if adequately pursued. Thus, in the production of electricity the coal station can compete favourably with the nuclear energy station, because of the greater inherent efficiency that has been developed over recent decades through improvements in technique and design.

The same story of change and advancement could apply to the gas industry, since the technical outlook of the industry has changed radically. It is not likely that the gas industry will build in the future conventional carbonizing plants, for attention is now being turned towards the total gasification of coal, bringing into use new techniques applicable to the use of cheaper low-rank coals. With these there may well be combined processes involving the gasification of oil, and the use of imported liquid methane. With high-pressure gas generation, pipe-line distribution over long distances becomes practicable and economic.

In regard to oil, the question of availability has been changed by the outlook regarding the discovery of new sources of oil in the Middle East, Libya and the Sahara, and of large natural gas fields in the Pyrenees. Against this must be set the likely growth of demand for energy, as the under-developed countries in the world respond to modern standards of living.

Sir Harold Hartley, in considering the energy outlook, has attempted to sketch the future pattern of energy consumption in Britain. Some 280 million tons of coal equivalent will be required per annum by 1965. Of this, some 9¼ million tons may come from nuclear energy, and 8 million tons in coal equivalents from hydro-electricity, coke and oil for generating power, leaving some 55 million tons per annum for steam generation by coal, an increase of some 10 million tons over 1959. The true forecast for the growth of other uses of coal, gas and oil offers a more difficult problem, and may well depend upon the pattern of technological advancement. Gas, provided it can be made at a competitive price, has potentialities in the domestic and industrial sphere, at three different levels of calorific value: a domestic gas at 500 or more B.T.U./cu. ft.; a synthesis or industrial gas of 320 B.T.U.; and a producer gas of 180 B.T.U./cu. ft., for which there are still uses, particularly in specialized fields and where the gas can be used, as in coke production, to free fuels of higher calorific value.

The competitive power of coal and its derived fuels can only be enhanced if the needed technological effort is put into the attempts to develop the new processes needed. Inevitably, the future demand will tend towards requiring energy either through a wire or a pipe, with any question of handling refuse or polluting the atmosphere excluded. Further, in the direction of the total gasification of coal lies the only hope of solving the problem of sulphur pollution.

In the matter of these potential fields of development an adequate tempo of research and development has been sadly lacking. Sir Harold voices the opinion that national policy in regard to research and development in the field of energy is badly out of balance. Very large sums of money and much available scientific effort are devoted to the nuclear field and insufficient of these resources to the utilization of coal, our greatest national asset.

R. J. SARJANT

HOST INFLUENCE ON PARASITE PHYSIOLOGY

THE sixteenth annual conference sponsored by the Bureau of Biological Research was held at Rutgers University, New Brunswick New Jersey, during January 29–30. The subject for discussion was host influence on parasite physiology.

The chair was occupied in turn by Dr. L. A. Stauber (Rutgers University), Dr. Norman Stoll (Rockefeller Institute) and Dr. Wm. Trager (Rockefeller Institute).

In his opening remarks, Dr. T. von Brand (Bethesda) observed that there was a time not very far back when a person interested in parasite physiology was regarded as an oddity. Now a 'physiological bandwagon' was emerging, possibly a reflexion of the brilliant advances being made in biochemistry. The study of parasite physiology, he considered, had changed in many ways; more people were interested and there was an emphasis on instrumentation, which demanded complicated apparatus, although man himself was the chief factor in any research. He recalled that during his youth in Germany, Weinland, that prince of experimenters, had been held up in his work on *Ascaris* metabolism for lack of a reducing valve. Distinct differences were apparent on the lines along which research on free-living invertebrates and parasites had developed. With the former the most significant advances were made by von Frisch in the field of sensory physiology. The study of parasites had developed along more chemical lines, possibly due to the fact that such early workers as Bunge and Weinland were interested primarily in chemical reactions. The biochemistry and physiology of parasites were initiated and extended in seeking to elucidate the mode of action of old or in the search for new drugs. Advances in our knowledge of host–parasite relationships followed. Non-biochemical studies of parasites were, however, of equal importance.

Dr. L. R. Cleveland (University of Georgia) then gave a paper on the effects of insect hormones on the protozoa of *Cryptocercus* and termites. This wood-feeding cockroach *Cryptocercus* has nine families, fourteen genera, and more than thirty species of flagellate parasites in its hind-gut. During each moulting period of a host all these species undergo sexual cycles. In the flagellates of adults and intermoult nymphal hosts, sexuality never occurs under natural conditions; when the insect growth and differentiation hormone, ecdysone, is injected into such hosts, gametogenesis is induced in their flagellates, and the time required for induction depends, within certain limits, on the amount of ecdysone injected. In an adult, 2,000 units will induce it in four genera within 3 hr.; 100 units will do the same in a 4th instar intermoult nymph.

Under natural conditions the sexual cycles of some genera of flagellates require six days, some nine, some twenty-six, and others 45–50 days for completion; 100 units of ecdysone, given to a 4th instar intermoult nymph, will bring all cycles to completion in seven days.

In intermoult nymphs and adults, injection of ecdysone produces sexuality in parasites without moulting of hosts.

Dr. J. D. Fulton (National Institute for Medical Research, London) spoke on some aspects of research on trypanosomes. The relationships of the three African trypanosomes, *T. brucei*, *T. gambiense* and *T. rhodesiense*, was first discussed. They cannot be distinguished morphologically and have sometimes been regarded not as separate species but as three biological races of a single species, *T. brucei*. Their distinguishing characters were dealt with in detail. The Tinde experiment started by Corson in 1934 had thrown considerable light on the affinities of these three parasites, on their physiological characters as well as on host–parasite relationships. The significance of polymorphism in trypanosomes and the presence of cytoplasmic inclusions, each character a facet of the host–parasite relationship, were discussed. The evidence suggested that sexual processes had not been demonstrated in these parasites. Dr. Fulton also described experiments on transformation in trypanosomes by means of deoxyribonucleic acid using drug resistance as a marker. The results obtained were negative or equivocal.

Dr. R. E. Hungate (University of California, Davis) described the factors influencing rumen protozoa. He showed how the ruminant provides to its symbiotic microbiota relatively constant temperature, moisture, osmotic pressure, *p*H, anaerobiosis, and food. The effect of variations in these factors on the protozoa was discussed, and the ability of the latter to withstand the normal variations in these factors was emphasized. In particular, the stress at the time of transfaunation was considered, including the devices for maintaining the necessary environment. The chief factor causing variation in the rumen microfauna is presumed to be differences in food. The effects of food may not be direct but mediated through bacteria which act upon it before they are consumed by the protozoa. The utilization of cellulose and the rapid ingestion of starch were stressed as characteristic of certain protozoa. The possible importance of protozoa during adaptation from hay to a high grain ration was presented.

Dr. A. R. Timms (Louisiana State University School of Medicine) dealt with the work of that School on schistosome enzymes. He explained how *Schistosoma mansoni* converts glucose quantitatively to lactic acid by the Embden–Meyerhof scheme of phosphorylating glycolysis which is also a major pathway for glucose utilization in mammalian tissues. However, enzymes from the parasite and the host, although catalysing the same biochemical transformations, are immunologically different.

Antimonials were shown by Bueding *et al.* to produce their antischistosomal activity, at least in part, by specific inhibition of phosphofructokinase activity of *S. mansoni*, leaving the host's phosphofructokinase largely uninhibited.

A protease was described which was thought to be in the gut of schistosomes, and which showed a marked specificity for hæmoglobin and globin when compared with other blood proteins. The possibility was discussed that this enzyme might provide an essential source of amino-acids for the parasite by degrading the host's hæmoglobin.

The paper by Dr. D. Fairbairn (McGill University, Canada) dealt with the physiology of hatching and larval exsheathment in nematodes. A result of investigations conducted in Canada and Australia has been that hatching and exsheathing of infective ascarid eggs and trichostrongyle larvæ can now be induced *in vitro* under conditions which closely resemble those in the host's intestinal tract. Under controlled conditions of *p*H, oxidation-reduction potential, temperature and concentration of free carbon dioxide, these processes occur at a rate comparable to that observed *in vivo*, and with identical morphological changes. Quantitative differences in the requirement of individual species for the components of this complex stimulus have been observed and positively correlated with conditions in the host. Exsheathment of *Haemonchus contortus*, for example, occurs only at high concentrations of free carbon dioxide such as are present in the sheep rumen, while a condition for optimal hatching of *Ascaris lumbricoides* eggs is that the carbon dioxide concentration be much lower, approximating that found in the small intestine of feeding mammals. The free-living infective eggs and larvæ cannot continue their development in the absence of the foregoing stimulus, which is in general peculiar to the host's intestinal tract, and thus ensures the transition to a parasitic existence. The processes of hatching and exsheathment constitute an internal response to stimulation, which is enzymic and results in digestion of the egg-shell and sheath.

The final paper, by Dr. P. P. Weinstein (Bethesda), was concerned with excretory mechanisms and excretory products of nematodes. The mechanism by which excretory products are released is poorly understood. Passage via cuticle, elimination by gut and so-called 'excretory system' may occur alone or in combination. This latter system was named primarily on teleological grounds, and little evidence for excretory function is available. Such a 'system' has been implicated in osmoregulatory activity and in moulting. Possible mechanisms for discharge of fluid such as secretion accompanied by preferential reabsorption were considered. The gut may play an important part in excretion of waste and the role of the cuticle and other structures was evaluated. The identification of the chemical nature of excretions and secretions was of possible importance regarding sensitization, development of functional immunity and pathological effects in the host. Products from axenically cultured parasitic nematodes are the only ones available for analyses. Much waste is apparently liberated as amino-acids and peptides or as organic acids, especially C_5 types. *In vitro* culture methods in defined media are urgently required for these studies.

A summary of the conference was given by Dr. Ernest Bueding (Louisiana State University) as follows. Since the occurrence of many biochemical reactions has been demonstrated in a large number of animal species of unicellular organisms and of plants, there has been a tendency to over-emphasize similarities and to overlook differences among various forms of life. Such biochemical differences may concern metabolic pathways and reactions (for example, fermentation products). In addition, it has been demonstrated that enzymes catalysing the same reactions in different species may not be identical with each other. Considerable biochemical differences have been uncovered not only between parasites and their hosts, but also among various parasitic species, even though they may be related to each other morphologically and taxonomically. Therefore, in biochemical studies of parasites, similarities and differences must receive equal attention and any one-sided view will result in a distorted picture. It is suggested that comparative biochemistry should be concerned with the nature of biochemical variability from patterns common to many forms of life; this, in turn, should contribute to a better understanding of biochemical evolution to a parasitic form of life.

J. D. FULTON

VIBRATIONS: THEIR EFFECTS AND ANALYSIS

IN this technological age, in which larger and larger amounts of power are being harnessed to aid mankind, it is not surprising that the unwanted adjunct of power, vibration, is becoming of great importance. Thus, for example, it is not only the public itself which is annoyed and fatigued by the noise of jet aircraft, but also the fabric of aeroplanes is now tending to show signs of fatigue and to crack. Again, in the field of supersonic transport, the motion of the aircraft can be of a frequency (around 5 c./s.) at which the body is least attuned to absorb the vibrational energy imposed upon it.

These topics, together with the more industrial problems of machinery vibration and occupational diseases arising from excessive vibration of drills, etc., were the subjects discussed at the recent meeting of the Acoustics Group of the Physical Society, held on January 20, at the Imperial College of Science and Technology, London.

Dealing first of all with problems of the vibration isolation of machinery, Mr. P. H. Allaway (Absorbit, Ltd.), in a review paper, gave an excellent appraisal of the many types of isolating materials and systems now available. The effect of the rigidity of the ground or floor structure was discussed, together with the effect of damping in shock vibration isolation. He emphasized the need for choosing the right type of installation for any type of application, and showed in the ensuing discussion a wealth of valuable experience and 'know-how'.

Dr. John Agate (Ipswich and East Suffolk Hospital) presented a somewhat frightening picture of the influence of vibrations in industry on the hands of operating personnel. Four diseases were mentioned in all: cysts of carpal bones, osteo-arthritis, soft-tissue injuries and a circulatory disturbance known as Raynaud's phenomenon. The small cysts can only be seen on X-ray examination and are not of any significance. Osteo-arthritis of the elbows is, of course, not confined to people working with vibrating machinery, though there is ample evidence of this being a contributory cause. Raynaud's phenomenon, which Dr. Agate dealt with extensively, is a circulatory disorder of the blood stream in the hands, and it arises from their operating in an environment of heavy vibration. In such cases, there is a lessening of the blood stream in the small arteries of the hands, and it is typified by the fingers and even the whole hand becoming white or blue. It is difficult to schedule the disease as a disability, since most fingers show these symptoms in the cold; further, many workers using vibrational types of machinery do not suffer with this loss of circulation in the fingers in the warmth of their factories, but only show the symptoms later in colder conditions, and then about six to eighteen months after first exposure.

The prevention of this disease is not easy, since any frequency less than 600 c./s. causes the damage to the arteries, the range of 40–125 c./s. nevertheless being the most serious. The lesson to be learnt is, in fact, the need for automation for any processes of this kind. Since this can only be introduced in very special instances, the solution is still unavailable and needs further scientific study.

In view of the recent interest in supersonic transport aircraft, Flight Lieut. Guignard's description of the work of the R.A.F. Institute of Aviation Medicine on physiological effects of low-frequency vibration was both interesting and significant from the point of view of design. The jet liners of the present have a natural frequency of oscillation of the fuselage structure somewhere in the region of 4–6 c./s., and pilots placed in the cockpits of these aircraft have been known to suffer from the up-and-down movements of that part of the long fuselage. The experiments described illustrate the reasons for this, and emphasize the need for designing away from such frequencies. It is found (Fig. 1) that if a male person, in a relaxed sitting posture, is vibrated vertically through the seat, a resonance occurs at some 5 c./s., the amplification of the head and shoulders being as much as three times that of the seat movement. Above this resonance frequency, the damping in the body eliminates harmonics of higher frequency. The effect of tensing the body is favourable, increasing the resonant frequency to 6·3 c./s. and also increasing the damping, thus reducing the amplitude of head movement by a small though significant amount. At these frequencies around 5 c./s., tests in the United States indicate a serious reduction in performance with poor eye-correlation, fatigue and pains in the chest.

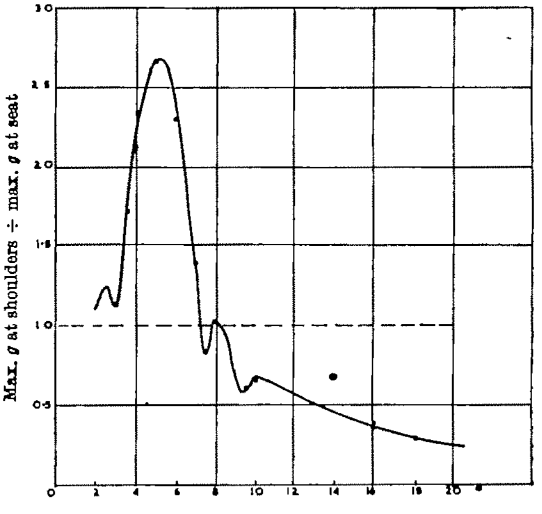

Fig. 1. Ratio of shoulder to seat acceleration plotted against frequency. Note the sharp resonance of the body at 5 c./s.

It is clear that this is a phenomenon of vital significance in the design of aircraft. The problem is not confined to aircraft, however. Ships cannot in any way claim freedom from vibrations of this frequency, while even trains, 'juddering' to a standstill in railway stations, set up exactly this type of disturbance.

Two complementary papers concerning the fatigue of aircraft structures due to vibrations set up by jet noise were presented respectively by Prof. E. J. Richards and Dr. B. L. Clarkson, of the Department of Aeronautics, University of Southampton. Jet noise arises from the turbulent motion of the jet gas as it mixes with the surrounding air. The total radiated energy is only a very small fraction of the energy of the jet, however (say, a hundredth of 1 per cent), and elaborate methods of experimentation have to be devised to measure the turbulence functions which are directly related to the radiated noise. In his paper, Prof. Richards described the space-time correlator and other apparatus used to obtain the spectrum of turbulence as seen by an observer moving with the jet stream. The relevant speeds of convection were discussed and a first attempt made to predict the radiated sound from the spectra of turbulence measured in this way.

There is, in general, no one discrete frequency of sound emitted from a jet. Consequently, the adjoining structure is vibrated randomly by the noise field and can in theory oscillate in an infinity of modes of oscillation. If a large number of modes do occur in practice, it is clear that the theoretical approach to prediction must be different from that adopted if the vibration is of a relatively simple kind. Dr. Clarkson's paper described experiments carried out on the vibrations of the rear part of the *Caravelle* airliner due to jet efflux, a programme carried out jointly by the firm, Sud-Aviation, and the University of Southampton. The analysis was concentrated on the strains in the centre of four panels of the fuselage skin aft of the engine nozzle; the first three being in line between two circumferential frames and the fourth being adjacent to the centre gauge of the first group but separated by a frame. Correlation measurements indicated that the lower frequencies (up to 500 c./s.) were associated with essentially overall vibration modes and have low strain amplitude. At the higher frequencies, having the larger strains, the frames tended to act as boundaries and the mode of vibration was confined to the stringers and panels between any two adjacent frames. In these measurements on this particular aircraft, the main resonance peak in each panel occurs at about 600–700 c./s. and has been identified with the fundamental stringer twisting mode (that is, adjacent panels are 180° out of phase). Strain response in the higher frequency range of 800–1,000 c./s. appears to be associated with a considerable number of closely spaced resonant frequencies.

This method of analysis using correlation techniques is a considerable step forward in the study of random vibrations of aircraft, ships and submarines, and the success outlined in the paper suggests that greater use should be made of this method of vibration analysis.

E. J. RICHARDS

A CONVECTIVE-TURBULENCE THEORY OF THUNDERSTORMS

By DR. N. RAMALINGAM

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IT is well known among meteorologists that in practical forecasting of thunderstorms the methods in current usage, such as the parcel methods, etc., of analysis of the dry-bulb and wet-bulb soundings of the atmosphere plotted on $T - \varphi$ grams or other thermodynamic diagrams, are often unreliable.

The initiation of thunderstorms, at least in such cases that occur in a calm atmosphere over land on hot afternoons, as experienced over India in the hot pre-monsoon months, has been investigated from the point of view of modern knowledge about turbulence. In doing that, however, the limiting condition:

$$\frac{\rho_1 - \rho_0}{\rho_0} > \frac{27\pi^4 k\nu}{4gh^3} \qquad (1)$$

given by Lord Rayleigh[1] from his mathematical investigation of the motion in the Bénard cells formed, when a fluid of thickness h in equilibrium with its density ρ_1 at the top being greater than its density ρ_0 at the bottom breaks down into convection, is taken as the main physical basis. In the above expression ν is the kinematic coefficient of viscosity, and k is the coefficient of conduction of heat of the fluid.

Indeed, even so far back as 1925, Brunt[2] made it very clear that some of the phenomena associated with thunderstorms can be explained in the same way, and that the violence of thunderstorms would appear to indicate that a high degree of instability is built up before the breakdown occurs. This suggestion, however, does not appear to have been pursued quantitatively.

In his original paper, Rayleigh gave the limiting condition as:

$$\beta' r > \frac{27\pi^4 k\nu}{4h^4} \qquad (2)$$

where $\beta'(= -\beta)$ is the lapse-rate; and $r = g\alpha$, α is the coefficient of expansion of the fluid, and g is the acceleration due to gravity.

Since he was considering a fluid of small thickness h, he took:

$$\beta' = -\beta = \frac{\theta_1 - \theta_0}{h}$$

θ being the departure from the equilibrium temperature, and:

$$r = g\alpha = g(\rho_1 - \rho_0)/\rho_0(\theta_1 - \theta_0)$$

hence, (2) reduces to (1).

If, however, we take a layer of air of thickness z, then we may take $\beta' = \dfrac{d\theta}{dz}$; and from the relation $\rho_1 = \rho(1 + \alpha d\theta)$ and taking $\rho_1 - \rho = d\rho$, we may write : $r = g\alpha = \frac{g}{\rho}\frac{d\rho}{d\theta}$, so that (2) reduces to :

$$\frac{d\rho}{\rho} > \frac{27\,\pi^4 k\nu}{4\,g\,z^4}\,dz \qquad (3)$$

Since we are dealing with atmospheric air where the heat is gained only at the bottom near the ground, the rest of the motion being adiabatic, the last condition becomes :

$$\frac{dP}{P} > \frac{27\,\pi^4\gamma k\nu}{4\,g\,z^4}\,dz \qquad (4)$$

where P is the atmospheric pressure, and γ is the well-known adiabatic coefficient for air (= 1·4). Since in the atmosphere, except very close to the ground, transmission of momentum and heat is brought about mainly by the turbulent eddies, the above limiting condition may be written as :

$$\frac{dP}{P} > \frac{27\,\pi^4\gamma K_M K_H}{4\,g\,z^4}\,.\,dz \qquad (5)$$

where K_M is the eddy viscosity and K_H is the eddy conductivity.

Since we are dealing with a large-scale phenomenon, such as the thunderstorm, much error may not arise, if, for simplicity, K_M is assumed to be nearly equal to K_H. Then from the expression for K_H quoted by Sutton[3], we may write :

$$K_H = l^2_H \sqrt{\left(-\frac{g}{\theta}\frac{d\theta}{dz}\right)}$$

where l_H is the mixing length and θ is the potential temperature. Further we take, with Prandtl, $l_H = \varkappa z$, where $\varkappa$ is the well-known von Kármán's constant (= 0·38). Then the limiting condition (5) reduces to :

$$d \log P > \frac{27\,\pi^4\gamma\varkappa^4}{4}\,(-\,d \log \theta) \qquad (6)$$

Summing up for all possible eddies, Σ of left-hand side must exceed Σ of the right-hand side. In other words, integrating from some level, l, to the ground-level, 0, we get the critical condition for the convective turbulent breakdown in the atmosphere as :

$$\frac{\log \dfrac{P_1}{P_0}}{\log \dfrac{\theta_0}{\theta_1}} > \frac{27}{4}\,\pi^4\gamma\varkappa^4 \qquad (7)$$

(taking $\varkappa = 0·38$, the right-hand side = 19·2 which is rounded off as 19. If $\varkappa$ is taken as = 0·37, the right-hand side could be taken as 17).

We therefore get the critical condition $\log \frac{P_1}{P_0} \Big/ \log \frac{\theta_0}{\theta_1}$ should exceed 19 (taking $\varkappa = 0·38$) for the breakdown in the equilibrium of the air to occur.

Since the left-hand side occurs as a ratio the logarithms may as well be taken to the base 10. Of course, to be more precise, one can take the potential temperatures corresponding to the two virtual temperatures. But this will not be of much significance. (Conversely, from a calculation of the left-hand side from the atmospheric sounding obtained before the occurrence of the thunderstorm, one can derive the value of von Kármán's constant by this independent method.)

For purposes of practical forecasting of thunderstorms while P_0 (about 1,000 millibars) refers to the ground-level, it will be sufficient if P_1 is taken as 500 or 600 millibars, since the lowest layer of 15,000 ft or so only will be of importance ; θ_0 and θ_1 are the potential temperatures corresponding to the dry-bulb temperatures at those levels.

Examination of a few random cases of the actual soundings of the atmosphere taken before thunderstorms seem to confirm the validity of the above critical condition (7). The same critical condition also appears to be applicable to other thunderstorms other than of the simple type assumed.

If the early morning sounding shows the ratio to just fall short of 19, one may be able to forecast whether a thunderstorm is possible later in the day by merely predicting whether insolation will be able to raise θ_0, so that the ratio exceeds 19, as the only other variable θ_1(P_1 and P_0 already having been chosen by us) is likely to remain practically the same, unless there is reason for believing that the air mass would have altered much in the few hours.

Of course, for a correct application of the critical condition (7), the sounding must strictly be obtained from a direct vertical ascent, as the soundings normally obtained by balloons are really 'spread over' observations.

In passing, it may be mentioned that while in the parcel method, etc., the $T - \varphi$ gram is used as an energy diagram, in this case the $T - \varphi$ gram serves little purpose except perhaps as a nomogram to read off the potential temperatures, and to judge the moisture content.

It follows, incidentally, from the above investigation that, surprisingly, the moisture in the air apparently seems to play a minor, or subsidiary, part so far as the initiation of the thunderstorm is concerned. Examination of the $T - \varphi$ grams referred to above also seem to confirm this view. This, perhaps, seems to be the reason why in the parcel method, thunderstorms are found to occur even in cases when the latent instability, as shown by the positive area, is little or nil ; and vice versa. Lest it should be misunderstood, it is clarified further. Water vapour being a constituent of the air, it also partakes fully in the convective processes of the air as outlined above ; and once the breakdown occurs in the equilibrium of the air, the water vapour will also undergo convection with its attendant condensation and release of latent energy, which all go to aggravate further and accelerate the vertical motions. If the moisture content of the air particularly at the lower levels is large, as shown by the wet-bulb temperatures being closer to the dry-bulb temperatures in the soundings, then when the breakdown occurs, there will be greater condensation and greater release of energy resulting in violent vertical motions that result in the formations of great vertical columns of cumulo-nimbus clouds, showers and all other attendant phenomena normally associated with thunderstorms. If the moisture content is less, and still the critical number is exceeded, and the breakdown occurs, there will probably be only development of a few cumulus clouds ; while if the air is practically dry, then something like what is known among aviators as 'clear air turbulence' will presumably result.

More details of this investigation will be published elsewhere.

[1] Rayleigh, Lord, *Phil. Mag.*, **32**, 529 (1916).
[2] Brunt, D., *Nature*, **115**, 299 (1925); "Physical and Dynamical Meteorology", 2nd ed., 219 (1939).
[3] Sutton, O. G., *Quart. J. Roy. Met Soc.*, **74**, 13 (1948)

VARIATION OF ELECTRON DENSITY IN THE IONOSPHERE WITH MAGNETIC DIP

By SHEILA CROOM, AUDREY ROBBINS and J. O. THOMAS

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THIS communication describes how the average ionospheric electron density (N_h) measured at local noon on quiet days in December 1957, at a series of fixed heights (h) in the F region, varies with the magnetic dip (I). The electron density profiles were deduced from observed $h'(f)$ curves by a method already described[1,2], the calculations having been carried out on the electronic digital computer, *Edsac II*, in the Mathematical Laboratory in the University of Cambridge.

It has been previously reported[3] that in September 1957 N_h varied with magnetic dip in an interesting and somewhat unexpected manner, and that the variation differed from that of the peak electron density N_mF2, particularly in the lower parts of the $F2$ layer. It was shown that, in these parts, the N_h (I) curve had maxima in N_h, where the dip was about 60° and there was evidence that the maxima occurred nearer the magnetic equator as greater heights were investigated.

Our previous investigations have now been extended to include the northern winter solstice, particularly in order to investigate whether N_h is abnormally large, corresponding to the well-known enhancement of N_mF2 described below.

When values of N_mF2 observed at different places are studied, two anomalies can be found: one, called the winter or seasonal anomaly, in which the values of N_mF2 are greater than expected on a simple theory in the winter hemisphere at medium and high latitudes, and the other, called the December or non-seasonal anomaly, in which, all over the Earth, the values measured in December are greater than expected. These variations have recently been studied in detail by Yonezawa[4]. At places in medium and high latitudes in the northern hemisphere, the two anomalies are in the same sense, so that the recorded values of N_mF2 are greater in December and January than at any other time of year.

Fig. 1 shows N_h at noon at a series of different heights plotted against magnetic dip for the month of December 1957, and may be compared with Fig. 1 in our previous communication[3], which was plotted for September 1957. The values plotted for N_h are the averages for all the good records for the ten International Quiet Days. The average values of N_h are estimated to be correct to within 5 per cent for stations in the northern hemisphere. The results for the southern hemisphere stations are probably somewhat less accurate. The observatories for which the points are plotted are listed in order of magnetic dip in Table 1.

The main features of the curves shown in Fig. 1 are the striking maxima in N_h, which occur at the larger values of h, in the form of a narrow peak centred on a dip of 70° (N.) and extending 10° on either side. The values of N_h at the 70° peak are probably exceptionally large as a result of the abnormally high sunspot number for this month ($\bar{R} = 239$).

Once it is realized that the winter anomaly in N_h may be restricted to a comparatively narrow

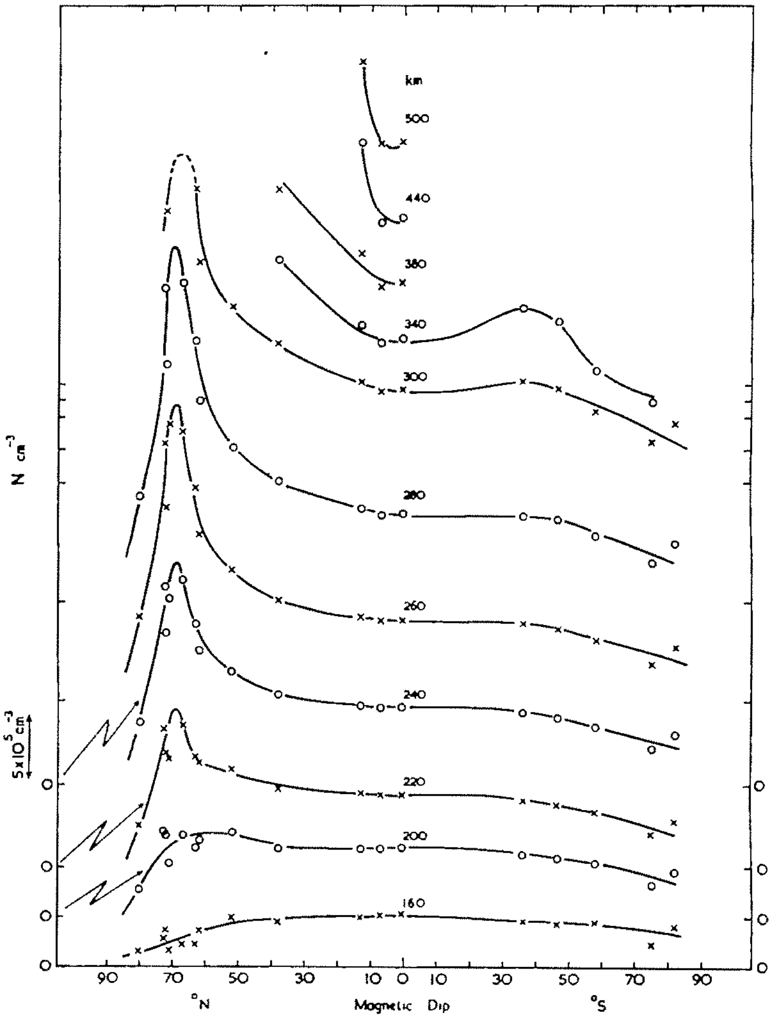

Fig. 1 Variation with magnetic dip of the mean quiet electron density N at a series of fixed heights in the F region at local noon in December 1957. The zero of the scale for the electron density at each height is marked. The stations used are listed in order of decreasing magnetic dip in Table 1

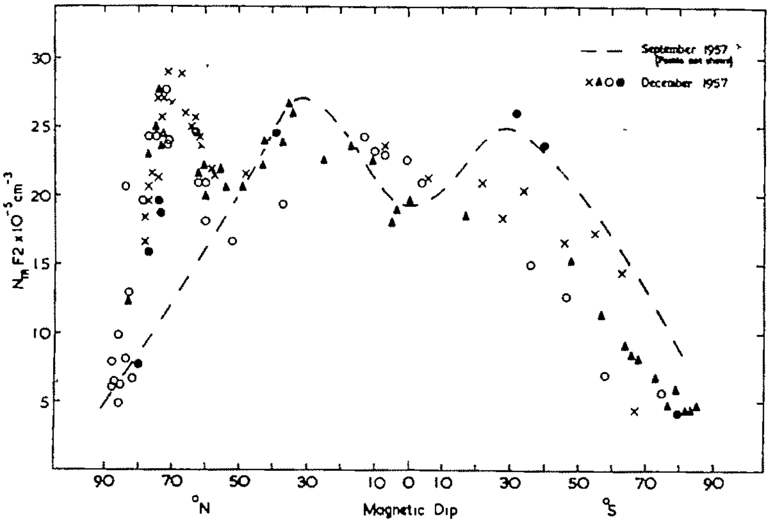

Fig. 2. Variation with magnetic dip of the monthly median noon values of N_mF2 in December 1957 for four geographical longitude zones. ×, 30° W.–50° E.; ▲, 50° E.–180° E.; ●, 180° W.–130° W.; ○, 130° W.–30° W. The broken line shows for comparison the variation of the monthly median N_mF2 for September 1957 (individual values are not shown)

range of dip angles, it is of interest to investigate how N_mF2, at the peak of the layer, depends on the dip in the same month. In Fig. 2 the monthly median values of N_mF2 for 12.00 L.M.T. are plotted against I, and it is at once evident that this quantity, also, exhibits the same comparatively narrow peak of increased ionization.

The monthly median values of N_mF2 have been separated in Fig. 2 into four main zones of geographical longitude. It is clear that the 70° peak in N_mF2 is independent of longitude. The broken line in Fig. 2 shows, for comparison, a smoothed curve of the variation of N_mF2 against dip in September 1957. This curve shows no indication of a peak near $I = 70°$ (N.). Results similar to those for December 1957 were obtained for the months of October and November 1957 and January 1958.

When values of N_mF2 are plotted against dip for other hours of the day and night, it is found that a similar maximum occurs near $I = 70°$ (N.) throughout the daylight hours. It appears at sunrise, is most prominent near noon and then decreases until at midnight it has disappeared.

Similar analyses carried out on observations for years with smaller sunspot numbers indicate that the size of the peak decreases with decreasing sunspot number.

A similar peak in N_mF2 occurs in the southern hemisphere in winter when the sunspot number is high. This is illustrated in Fig. 3, where a smooth curve has been drawn through the monthly median values of N_mF2 for July, September and December 1957.

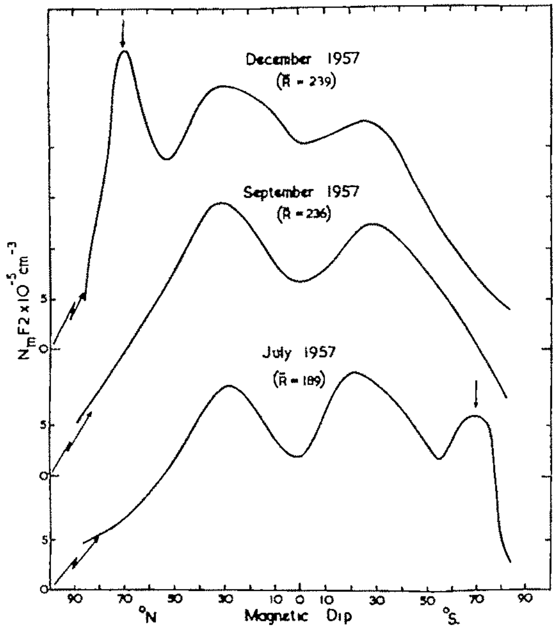

Fig. 3. Variation with magnetic dip of monthly median N_mF2 at local noon in July, September and December 1957. $\bar{R}$ is the monthly average relative Zurich sunspot number

A comparison of the variation of N_h with dip in December 1957 (Fig. 1) with the variation of N_mF2 for the same month (Fig. 2) indicates that, below about 300 km., there is no evidence in N_h of the minimum which occurs at 50° (N.) in N_mF2, and that N_h in this month decreases uniformly towards the magnetic equator from a dip of 60° (N.), as it did in September 1957 (Fig. 1 of ref. 3).

Table 1. DETAILS OF STATIONS FOR WHICH $N(t)$ DATA WERE AVAILABLE
$N(h)$ profiles for stations marked *C* were computed at the Cavendish Laboratory. The results for stations marked *S* were taken from ref. 5

Station	Source	Geographical latitude	Geographical longitude	Magnetic dip
Point Barrow	*C*	71° 08′ N.	156° 48′ W.	80° N.
St. John's	*C*	47° 33′ N.	52° 40′ W.	72° N.
Washington	*S*	38° 44′ N.	77° 08′ W.	72° N.
Inverness	*C*	57° 27′ N.	4° 15′ W.	71° N.
Slough	*C*	51° 30′ N.	0° 36′ W.	67° N.
Adak	*C*	51° 54′ N.	176° 39′ W.	63° N.
San Francisco	*C*	37° 26′ N.	122° 10′ W.	62° N.
Puerto Rico	*C*	18° 30′ N.	67° 12′ W.	52° N.
Panama	*S*	09° 23′ N.	79° 53′ W.	39° N.
Talara	*S*	04° 34′ S.	81° 15′ W.	13° N.
Chimbote	*C*	09° 04′ S.	78° 35′ W.	07° N.
Huancayo	*S*	12° 03′ S.	75° 20′ W.	01° N.
Concepcion	*C*	36° 35′ S.	72° 59′ W.	36° S.
Port Stanley	*C*	51° 42′ S.	57° 52′ W.	47° S.
Port Lockroy	*C*	64° 50′ S.	63° 31′ W.	58° S.
Pole Station	*C*	90° 00′ S.		75° S.
Wilkes	*C*	66° 25′ S.	110° 27′ E.	82° S.

We are grateful to the Director of the Mathematical Laboratory of the University of Cambridge for permission to use *Edsac II* and to all the organizations, particularly the World Data Center *A*, Boulder, Colorado, U.S.A., which have lent us $h'(f)$ records. We should like to acknowledge the assistance of the computing staff of the Radio Group of the Cavendish Laboratory. This work was carried out with the aid of a grant from the Department of Scientific and Industrial Research.

[1] Thomas, J. O., Haselgrove, J., and Robbins, A. R., *J. Atmos. Terr. Phys.*, 12, 46 (1958).

[2] Thomas, J. O., and Vickers, M. D., D.S.I.R. Radio Research Special Report No. 28 (H.M. Stat. Office, London, 1959).

[3] Croom, S. A., Robbins, A. R., and Thomas, J. O., *Nature*, 184, 2003 (1959).

[4] Yonezawa, T., *J. Radio Res. Lab.*, 6, 651 (1959).

[5] Schmerling, E. R., "Tables of Ionospheric Electron Densities" (Penn. State University, 1957–58).

SIGNIFICANCE OF RIBONUCLEIC ACID IN THE MECHANISM OF FORMALDEHYDE-INDUCED MUTAGENESIS

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THE present article reports the significant occurrence that formaldehyde exhibits no mutagenic activity towards *Drosophila melanogaster*, by the larval feeding method, unless ribonucleic acid is present in the treatment medium.

In a study of the mode of action of chemical mutagens, the use of a chemically defined and axenic treatment medium is clearly essential for the precise definition and discrimination of both qualitative and quantitative effects. Such a medium has now been developed for *Drosophila melanogaster* by Sang[1], in which the dose-response for the main dietary constituents on the duration of larval life has been determined. On the basis of these findings, he has formulated an optimal medium (Table 1), which allows larval development to be completed in approximately 4·4 days ; this compares with 4·1 days when the same strain is allowed to develop on an optimal supply of killed yeast. The omission of ribonucleic acid from the synthetic medium prolongs the period of larval development to about seven days[1]. Thus, although *Drosophila* larvæ can normally develop without an exogenous supply of ribonucleic acid, its addition accelerates development, indicating that preformed purines (or their nucleosides or nucleotides) are assimilated, to supplement or replace the natural synthetic process.

Table 1. COMPOSITION OF STANDARD CULTURE MEDIUM

	(gm.)		(gm.)
Yeast ribonucleic acid	0·4	Na$_2$HPO$_4$	0·189
Casein ('Genatosan-low vitamin')	5·00	Aneurin	0·0002
		Riboflavine	0·0010
Fructose	0·75	Nicotinic acid	0·0012
Cholesterol	0·03	Calcium pantothenate	0·0016
Lecithin	0·40	Pyridoxine	0·00025
Agar ('Oxoid-Kobe No. 1')	3·00	Biotin	0·000016
NaHCO$_3$	0·14	Folic acid	0·0010
KH$_2$PO$_4$	0·183	Water to 100 ml.	

The folic acid content of this medium is about ten times the minimum required in a medium containing ribonucleic acid, since it is known that the folic acid requirement is increased when ribonucleic acid is omitted from the diet. This excess does not interfere with development when optimal ribonucleic acid is supplied in the food.

The *Drosophila* stock used in the present study, and in previous experiments[3,4], the Oregon-*K* strain, does, however, require a dietary supply of cytidine for normal growth and development[2] ; this was supplied as cytidylate (0·025 per cent). Since ribonucleic acid contains cytidylate, this was not added when ribonucleic acid was present. The medium will be referred to as the synthetic medium (*sm*) when ribonucleic acid is omitted, and as *sm* + ribonucleic acid when the acid is present. Ordinary medium is the yeast–maize meal–treacle medium normally used for culturing *Drosophila* in this laboratory. It was decided to omit the egg sterilization procedure and autoclaving of the medium normally essential with this technique, since formaldehyde itself effectively suppresses microbial growth, and propionic acid is used as a fungicidal agent, as in previous experiments. The procedure for treatment of larvæ and brood pattern determinations for sex-linked lethal mutation rates is as previously described[3]. The age of larvæ is counted from the time of hatching from the egg. The larval developmental time for treated larvæ was counted from emergence from the egg to puparium formation. In most experiments the larval developmental time is compounded of pre-treatment, treatment and post-treatment times.

It was decided to test the mutagenic activity of formaldehyde in the presence and in the absence of ribonucleic acid, now made possible by Sang's refined culturing technique.

Experiments were carried out at a constant concentration of formaldehyde ($2 \cdot 0 \times 10^{-2}$ *M*) with different levels of ribonucleic acid in the treatment medium. The optimal concentration of the acid in the synthetic medium has been shown by Sang to lie between 0·4 and 0·7 per cent[1]. Table 2 shows the results obtained over the concentration range 0·2–0·6 per cent ribonucleic acid, where larvæ were kept on the treatment medium for the whole of larval life. The mutagenic activity of formaldehyde in the first brood is seen to be significantly lower at the lowest concentration of ribonucleic acid tested (0·2 per cent) than at the 0·3 per cent ribonucleic acid level ($\chi^2 = 15 \cdot 36$ with one degree of freedom). The mutation frequencies of the first broods over the range 0·3–0·6 per cent ribonucleic acid are not significantly different from one another (heterogeneity $\chi^2 = 0 \cdot 18$ with three degrees of freedom). Second broods are sampled as a check against possible changes in brood patterns by influences on the speed of germ-cell development ; these are seen to be not significantly different from one another over the ribonucleic acid concentration range tested (heterogeneity $\chi^2 = 1 \cdot 98$ with four degrees of freedom). It should be noted that in the formaldehyde-containing synthetic medium (at least, at the concentration used here), the larvæ do not

Table 2. EFFECT OF THE CONCENTRATION OF RIBONUCLEIC ACID ON THE MUTAGENIC ACTIVITY OF FORMALDEHYDE, WHEN TREATMENT IS FOR THE ENTIRE LARVAL LIFE

Formaldehyde concentration	$2 \cdot 0 \times 10^{-2}$ *M*				
Ribonucleic acid concentration (per cent)	0·2	0·3	0·4	0·5	0·6
Larval developmental time (days)	6·5	6·0	5·5	6·5	7·5
Per cent survivors	75	80	58	54·5	65
	First broods				
No. chromosomes examined	602	598	632	336	326
No. lethals	18	54	54	26	38
Per cent lethals	2·99	9·03	8·54	7·74	11·65
	Second broods				
No. chromosomes examined	604	514	770	602	539
No. lethals	10	14	18	10	10
Per cent sex-linked recessive lethal mutations	1·65	2·72	2·84	1·66	1·85

Table 3. EFFECT OF RIBONUCLEIC ACID ON THE MUTAGENIC ACTIVITY OF FORMALDEHYDE, WHEN 18-HR. LARVÆ ARE TREATED FOR 24 HR.

Treatment medium	*sm*	*sm* + RNA (0·4 per cent) delayed	*sm* + RNA (0·4 per cent)
Formaldehyde concentration		$2 \cdot 0 \times 10^{-2}$ *M*	
Larval developmental time (days)	5·5	5·5	5·0
Per cent survivors	91·8	79·6	83·7
	First broods		
No. chromosomes examined	548	481	732
No. lethals	2	8	23
Per cent sex-linked recessive lethal mutations	0·36	1·66	3·14
	Second broods		
No. chromosomes examined	503	550	477
No. lethals	0	7	2
Per cent sex-linked recessive lethal mutations	0·00	1·27	0·42

survive if the ribonucleic acid concentration is less than 0·2 per cent or greater than 0·7 per cent.

While it was not possible to test the mutagenic activity of formaldehyde in the absence of ribonucleic acid, by treating larvæ throughout larval life, this difficulty was overcome after consideration of the following observations.

In a study of the response of *Drosophila* larval germ cells to the mutagenic activity of orally administered formaldehyde, Auerbach[5] has determined the patterns of sensitivity for each of the four days of larval life. In her initial experiments, larvæ were put on the formaldehyde food at the beginning of the stage to be treated, and transferred to ordinary food after 24 hr. It was afterwards discovered that, as formaldehyde food on which the larvæ are feeding loses its mutagenic activity within 24–48 hr.[5], transfer to ordinary food is not necessary; this means that approximately the same sensitivity pattern is obtained if the larvæ are allowed to complete development on the formaldehyde food as the ones transferred after only 24 hr. of treatment[5]. The ineffectiveness of formaldehyde food after 24–48 hr. does not appear to be due completely to loss of mutagenic activity of the medium[6], but rather to the development of a tolerance in treated larvæ which makes them incapable of responding to even a slightly deteriorated formaldehyde medium. However, if the larvæ which have spent some time on treated food are transferred to freshly treated food, formaldehyde can produce mutations in cells which would have failed to mutate with continued exposure to the old medium[7]. Observations on the sensitivity pattern of response of larval germ cells suggest the coincidence of the sensitive period with the auxocyte stage in primary spermatocyte development[6], that is, the protracted growth stage which precedes meiosis in the larval testis. Auxocytes are found to make their first appearance about the twelfth hour of the first day of larval life, which coincides with the beginning of sensitivity to formaldehyde food. Consequently, treatment of larvæ during the first day of larval life, whether the larvæ are allowed to complete development on the treatment medium, or are transferred to ordinary food after 24 hr., involves approximately the same effective treatment. In view of this latter consideration, it may be reasoned that the experiments quoted in Table 2 simply mean that, at the lowest concentration of ribonucleic acid (0·2 per cent), the appearance of the sensitive stage is delayed until the formaldehyde food has almost lost its effectiveness. This argument is unlikely, however, as the developmental time at 0·2 per cent ribonucleic acid is about the same as, and in one case (0·6 per cent) less than, for acid concentrations giving maximum mutagenic effect. Moreover, differences in speed of larval development do not necessarily result in altered mutational response to formaldehyde[7], although decreases in the mutagenic activity of formaldehyde have been reported after pronounced increases in the duration of larval life[7]. Nevertheless, it was decided to attempt to slow down development slightly on the synthetic treatment medium containing ribonucleic acid to obtain a comparable developmental time with the treatment medium where ribonucleic acid is omitted. This was achieved by halving the cholesterol concentration and substituting 0·004 per cent choline for the lecithin; these constituents were chosen as they have no known connexion with nucleic acid metabolism. Such a medium will be referred to as '*sm* + ribonucleic acid delayed'.

In consequence of the preceding observations, it was decided to pre-feed larvæ on *sm* + ribonucleic acid (0·4 per cent) medium for some time before transfer to the treatment medium, thus assuring that sensitive cells are present at the time of treatment. This procedure, however, has the disadvantage that a proportion of sensitive cells normally affected in the first brood will have passed the sensitive stage at the time of treatment; consequently, lower mutation rates occur.

Table 3 summarizes the results obtained when 18-hr. larvæ are treated for 24 hr., after which they are allowed to complete development on ordinary medium. In the absence of ribonucleic acid, no formaldehyde-induced mutagenic activity is detected; the spontaneous mutation-rate of the Oregon-*K* strain

Table 4. EFFECT OF RIBONUCLEIC ACID ON THE MUTAGENIC ACTIVITY OF FORMALDEHYDE, WHEN 24-HR. LARVÆ ARE TREATED FOR 24 HR.

Treatment medium	*sm*	*sm* + RNA (0·4 per cent)	*sm* + RNA (0·4 per cent) delayed	*sm* + RNA (0·1 per cent)	*sm* + DNA (0·4 per cent)
Formaldehyde concentration			$2 \cdot 0 \times 10^{-2}$ *M*		
Larval developmental time (days)	5·5	5·0	5·5	5·5	5·5
Per cent survivors	61·8	62·6	55·4	70·6	64·6
		First broods			
No. chromosomes examined	579	602	605	599	582
No. lethals	2	21	12	1	0
Per cent sex-linked recessive lethal mutations	0·34	3·5	1·98	0·16	0·0
		Second broods			
No chromosomes examined	412	370	402	432	418
No. lethals	1	2	4	1	0
Per cent sex-linked recessive lethal mutations	0·24	0·54	1·0	0·23	0·0

Table 5. EFFECT OF RIBONUCLEIC ACID ON THE MUTAGENIC ACTIVITY OF FORMALDEHYDE, WHEN 15-HR. LARVÆ ARE TREATED FOR 3 DAYS

Treatment medium	*sm*	*sm* + RNA (0·4 per cent)	*sm* + RNA (0·4 per cent) delayed
Formaldehyde concentration		2·0 × 10^{-2} *M*	
Larval developmental time (days)	6·5	5·5	6·5
Per cent survivors	> 70	> 70	> 70
	First broods		
No. chromosomes examined	608	501	557
No. lethals	0	28	21
Per cent sex-linked recessive lethal mutations	0 0	5·6	3·8
	Second broods		
No. chromosomes examined	352	367	354
No. lethals	0	5	3
Per cent sex-linked recessive lethal mutations	0·0	1·36	0·85

varies normally from 0·1 to 0·3 per cent recessive sex-linked lethal mutations. In both acid series there is significant mutagenic activity, and although '*sm* + ribonucleic acid delayed' has the same developmental time as '*sm*', it shows no significantly different mutagenic response from '*sm* + ribonucleic acid' in either the first or second brood (χ^2 = 2·5 and 2·14, with one degree of freedom, respectively).

A comparable experiment where 24-hr. larvæ were treated by the same procedure is shown in Table 4. Again, no induced mutations are detected in the absence of ribonucleic acid, whereas activity is significant in both the '*sm* + ribonucleic acid' and '*sm* + ribonucleic acid delayed'; the mutational response of the latter two treatments do not differ significantly from one another in either the first or second brood (χ^2 = 2·57 and 0·52 with one degree of freedom, respectively). In this series, treatments were also carried out where the concentration of ribonucleic acid was reduced to 0·1 per cent (*sm* + ribonucleic acid (0·1 per cent)), and one where 0·4 per cent thymus gland deoxyribose nucleic acid replaces ribonucleic acid (*sm* + deoxyribonucleic acid (0·4 per cent)): no significant activity was detected after either treatment. Deoxyribonucleic acid was used here as it is known not to be utilized by *Drosophila* larvæ, and is even toxic to them[8]; at the concentration used here, however, the toxic effect is negligible.

Table 6. EFFECT OF RIBONUCLEIC ACID ON THE MUTAGENIC ACTIVITY OF FORMALDEHYDE, WHEN 15-HR. LARVÆ ARE TREATED FOR 2 DAYS, AND RE-TREATED ON FRESH FORMALDEHYDE FOOD FOR A FURTHER 2 DAYS

Treatment medium	*sm*	*sm* + RNA (0 4 per cent)	*sm* + RNA (0·4 per cent) delayed
Formaldehyde concentration		2 × 10^{-2} *M*	
Larval developmental time (days)	7 0	6 0	6 8
Per cent survivors	70·8	45·4	55 0
	First broods		
No. chromosomes examined	744	640	668
No. lethals	2	16	22
Per cent lethals	0·27	2·5	3 3
	Second broods		
No. chromosomes examined	355	362	337
No. lethals	1	7	6
Per cent sex-linked recessive lethal mutations	0 28	1·93	1·78

A further series (Table 5) is summarized, where 15-hr. larvæ were left on the treatment medium for three days, after which they were allowed to complete development on ordinary food. Once more, no induced mutations are detectable in the absence of ribonucleic acid, whereas '*sm* + ribonucleic acid' and '*sm* + ribonucleic acid delayed' both show high mutation-rates which are not significantly different from one another in either the first or second brood (χ^2 = 1·98 and 0·43 with one degree of freedom, respectively). The developmental times of '*sm*' and '*sm* + ribonucleic acid delayed' were almost identical, despite the increased duration of the treatment.

A final series was carried out, where advantage was taken of the fact that if larvæ, which have spent some time on formaldehyde-treated food, are transferred to freshly treated food, formaldehyde can produce mutations in cells which would have failed to mutate with continued exposure to the old medium[7]. Daily transfers to fresh formaldehyde food were carried out, but the effect on the development was found to be too pronounced except at low (0·5 × 10^{-2} *M*) formaldehyde concentrations. However, less frequent transfers proved successful. Table 6 shows the results obtained when 15-hr. larvæ were left on formaldehyde food for two days, followed by transfer to fresh formaldehyde food for a further two days, after which they were allowed to complete development on ordinary food. No induced mutagenic activity was detectable in the absence of the acid, whereas both ribonucleic acid treatments showed mutagenic activity. The two acid treatments are not significantly different from one another in either the first or second brood (χ^2 = 0·73 and 0·02, with one degree of freedom, respectively); the developmental times of '*sm*' and '*sm* + ribonucleic acid delayed' were approximately the same.

Table 7. EFFECT OF RIBONUCLEIC ACID ON THE MUTAGENIC ACTIVITY OF URETHANE, WHEN TREATMENT IS FOR THE ENTIRE LARVAL LIFE

Treatment medium	*sm*	*sm* + RNA (0·4 per cent)
Urethane concentration	0·1 per cent	
Total developmental time	16–19 days	10–14 days
Per cent survivors	49	67·5
	First broods	
No. chromosomes examined	964	648
No. lethals	14	10
Per cent sex-linked recessive lethal mutations	1·45	1 54
	Second broods	
No. chromosomes examined	892	1,087
No. lethals	12	9
Per cent sex-linked recessive lethal mutations	1·34	0 83

That this phenomenon is not a general one for chemical mutagens is seen from Table 7, where I tested the mutagenic activity of urethane by the larval feeding method. Newly hatched larvæ were allowed to develop throughout larval life on the synthetic treatment medium under axenic conditions, and the mutagenic activity of urethane was tested in the presence and absence of ribonucleic acid; for both treatments urethane was autoclaved in the medium. Since urethane prolongs the pupal period, and adult emergence time is scattered over several days, developmental times are given from hatching from the egg to peak adult emergence. No difference was observed between the mutational response of the two series (first and second brood χ^2 = 0·275 and 1·25, respectively).

An analysis of the mechanism by which ribonucleic acid mediates the mutagenic activity of formaldehyde is now in preparation for publication.

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[1] Sang, J. H., *Proc. Roy. Soc. Edin.*, 67, 339 (1957).
[2] Ellis, J. F., *Phys. Z.*, 32, 29 (1959).
[3] Alderson, T., *Nature*, 179, 97 (1957).
[4] Alderson, T., *Nature*, 182, 508 (1958).
[5] Auerbach, C., and Moser, H., *Z. Vererbungslehre*, 85, 479 (1953).
[6] Barker, J. S. F., and Davern, C. I., *Aust. J. Biol. Sci.*, 9, 382 (1956).
[7] Auerbach, C., *Z. Vererbungslehre*, 9, 382 (1956).
[8] Villee, C. A., and Bissell, H. B., *J. Biol. Chem.*, 172, 59 (1948).

EFFECTS OF STARVATION ON THE ABILITY OF ISOLATED RAT DIAPHRAGM TO INCORPORATE LABELLED AMINO-ACIDS INTO ITS PROTEIN AND TO RESPOND TO THE INFLUENCE OF INSULIN ON THIS PROCESS

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IT is now well established that insulin *in vitro* stimulates the incorporation of labelled amino-acids into the protein of isolated rat diaphragm[1-4], although the mechanism by which this action is brought about remains unknown. The earlier experiments of Krahl[2] seemed to suggest that insulin enhanced incorporation of amino-acid into protein by favouring carbohydrate utilization, a process which would make available more energy and possibly more pentose for the synthesis of ribonucleic acid and protein. Addition of glucose to the medium, in place of insulin, could also simulate incorporation. More recently, both Manchester and Young[3] and Wool and Krahl[4] have described conditions under which addition of glucose has no observable effect on incorporation and yet where addition of insulin produces a very marked and consistent stimulation. However, Wool and Krahl[4] also find that a stimulating effect of glucose on incorporation of amino-acid can be observed with diaphragm from rats which have starved for 48 hr., the rate of incorporation in the absence of glucose being markedly depressed by comparison with diaphragm from the non-fasting animal. Under these conditions the effect of insulin on incorporation is very severely reduced. Similar results have not been obtained in this laboratory, and in the present communication evidence is presented to the effect that the nutritional status of the diaphragm donor (that is, the length of time, up to 48 hr., that the animal has starved before removal of the diaphragm) does not seriously modify the effect of insulin on incorporation, nor can the action of insulin on amino-acid incorporation be simulated in any way by glucose.

Diaphragm was obtained from female rats of either the albino Wistar or the black-and-white hooded strain, weighing about 120 gm. and fed a stock laboratory diet[6]. Diabetic animals had been rendered diabetic by subcutaneous injection of alloxan (200 mgm./kgm.) at least two weeks before removal of the diaphragm. Such animals had non-fasting blood sugar-levels above 500 mgm. per cent, but after the withdrawal of food for 24 hr. the blood sugar-level dropped to an average of about 215 mgm. per cent. Hemidiaphragms were incubated for 2 hr. at 37° C. in a bicarbonate buffer[7] *p*H 7·4, gassed with oxygen plus carbon dioxide (95 : 5) in the presence of 1–^{14}C–glycine (1 μmole/ml.), with or without addition of glucose (2·5 mgm./ml.) and/or insulin (Boots crystalline insulin – 22 units/mgm., 0·1 unit/ml.). The procedure for the removal and incubation of the diaphragm and for the preparation of protein samples has been previously described[5]. In the experiments with diaphragm from Wistar rats the glycine added to the medium had a specific activity of about 400 μc./m.mole; the protein prepared from diaphragms was collected on 1-cm.^{2} plastic disks as infinitely thick samples[8]. In the experiments with diaphragm from the hooded rats the glycine added to the medium had a specific activity of about 80 μc./m.mole; the diaphragm protein was collected as a thin film on 7·5 cm.^{2} aluminium disks, and its content of radioactivity assessed in counting equipment with an efficiency for carbon-14 of about three times that of the equipment used for counting the infinitely thick protein samples. The figures shown for incorporation of radioactivity into protein of diaphragm from the two strains of rat are therefore not comparable. Other experiments in which the rate of incorporation by diaphragm of the two strains has been compared suggest that the respective rates are fairly similar.

In the absence of insulin, incorporation of ^{14}C-glycine into the protein of diaphragm from Wistar and hooded rats starved 24 hr. was slightly (13 and 12 per cent) reduced by comparison with diaphragm from non-fasting animals (Table 1). A further 24 hr. of starvation did not appreciably reduce incorporation in either strain. In the presence of insulin, incorporation into protein of diaphragm from Wistar rats starved for 24 hr. had declined 10 per cent by comparison with diaphragm from non-fasting animals, but incorporation did not decline further when the diaphragm donors starved for 48 hr. (Table 1). Starving hooded animals for 24 hr. did not depress the rate of incorporation of glycine by the isolated diaphragm in the presence of insulin, though with diaphragm from rats starved for 48 hr., incorporation was appreciably (21 per cent) lower (Table 1). With diaphragm from the Wistar rats the magnitude of the effect of insulin on incorporation of ^{14}C-glycine was unchanged by the nutritional pre-treatment of the diaphragm donor, and was about four times the size of the change induced by starvation (Table 1). With

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* Mr. and Mrs. John Jaffé Donation Student, Royal Society.

Table 1. EFFECT OF FASTING ON THE INCORPORATION OF ^{14}C-GLYCINE INTO THE PROTEIN OF ISOLATED RAT DIAPHRAGM AND THE STIMULATION BY INSULIN OF THIS PROCESS

Strain of rat from which diaphragm taken	Food withheld (hr.)	Radioactivity in diaphragm protein		
		No insulin added	Insulin added (0·1 unit/ml.)	Difference
		(counts/min./disk)		
Wistar	0	281 ± 16 } $P < 0·1$	442 ± 19	161
	24	242 ± 14 }	406 ± 17	164
	48	250 ± 9	401 ± 16	151
		(counts/min./mgm.)		
Hooded	0	11·9 ± 0·94 }	18·0 ± 1·22 } $P < 0·05$	6·1
	24	10·4 ± 0·88 } $P < 0·1$	17·6 ± 1·56 } $P < 0·1$	7·2
	48	9·8 ± 0·62 }	14·2 ± 0·85 }	4·4

Each figure is the mean ± S.E. of the mean of six observations. The value of P for a difference which is significant is indicated. The effect of insulin is significant in each instance ($P < 0·01$). No glucose was added to the medium.

diaphragm from the hooded strain the size of the insulin effect was somewhat reduced when the donor had starved for 48 hr., though this diminution was not apparent with diaphragm from rats starved for 24 hr. (Table 1). The magnitude of the insulin effect, however, was still twice the diminution in incorporation produced by starvation.

Addition of glucose or pyruvate to the medium in either the presence or absence of insulin has previously been shown to have no observable effect on the incorporation of a large number of different ^{14}C-labelled amino-acids into the protein of diaphragm from Wistar rats starved for 20–24 hr.[3]. Addition of glucose to the incubation medium, again in the presence or absence of insulin, has now been found to have no appreciable or significant effect on the incorporation of ^{14}C-glycine into the protein of diaphragm from rats of either strain starved for 48 hr. or from alloxan-diabetic Wistar rats starved for 24 hr. (Table 2). The magnitude of the insulin effect is also unchanged by the addition of glucose, and is many times larger than any effect of glucose.

Since Wool and Krahl[4] were using rats of the Sprague-Dawley strain, the results presented here may indicate that there are quite large quantitative differences in the response of different strains to the effects of starvation, glucose and insulin on amino-acid incorporation. In particular, rats of the Sprague-Dawley strain, as judged by the ability of the isolated diaphragm to incorporate labelled amino-acids into protein, appear to be incapacitated to a greater extent by starvation than are those of the Wistar and hooded strains. In other words, availability of the energy required for protein synthesis becomes a rate-limiting factor at an earlier stage during starvation in the Sprague-Dawley rat. Perhaps because of this an enhancement of the rate of amino-acid incorporation by addition of glucose is more readily observed with the isolated diaphragm of this strain[5]. On the other hand, slight variations in the conditions under which the animals are reared and kept may lead to quite large differences in the response of diaphragm to the conditions investigated.

The results presented here, taken in conjunction with earlier papers from this laboratory[3,9], do indicate, perhaps more clearly than hitherto, that insulin *in vitro* invariably stimulates incorporation of labelled amino-acids into the protein of isolated diaphragm from rats in varied nutritional states, and that this effect of insulin is not dependent upon an effect of insulin on glucose uptake, neither can it be simulated by glucose alone, nor is it observably modified by the simultaneous presence of glucose. The availability of material acting as a source of energy for protein synthesis, even for the alloxan-diabetic animal, is not therefore the rate-limiting step in amino-acid incorporation in diaphragm, and the action of insulin on this process does not appear to be brought about by its action on carbohydrate metabolism. The slightly reduced basal incorporation observed here which results from starvation for 24 or 48 hr. may be a consequence of a reduced rate of secretion and lower circulating level of insulin than is the case in the non-fasting animal.

Although insulin enhances the rate of accumulation of a non-utilizable amino-acid, α-amino isobutyric acid, by diaphragm[10], it has not so far proved possible to show an effect of insulin on the accumulation of naturally occurring amino-acids in the same way[11]. The bulk of the evidence at the present time[11-13] is against the view that insulin enhances amino-acid incorporation into muscle protein by affecting the transfer of amino-acids into the cell, but the details of the mechanism by which insulin regulates amino-

Table 2. EFFECT OF THE ADDITION OF GLUCOSE TO THE MEDIUM ON THE INCORPORATION OF ^{14}C-GLYCINE INTO THE PROTEIN OF DIAPHRAGM FROM FASTING RATS AND THE STIMULATION BY INSULIN OF THIS PROCESS

Food withheld (hr.)	Strain of rat from which diaphragm taken	Addition to the medium	Radioactivity in diaphragm protein		
			No insulin added	Insulin added (0·1 unit/ml.)	Difference
				(counts/min./disk)	
48	Wistar	No addition	187 ± 8*	317 ± 11	130
		Glucose (2·5 mgm./ml.)	208 ± 9	312 ± 14	104
				(counts/min./mgm.)	
48	Hooded	No addition	10·7 ± 0·68	16·7 ± 1·29	6·0
		Glucose (2·5 mgm./ml.)	11·7 ± 0·81	17·5 ± 0·82	5·8
				(counts/min./disk)	
24	Alloxan-diabetic Wistar	No addition	200 ± 14*	337 ± 23	137
		Glucose (2·5 mgm./ml.)	203 ± 11	338 ± 21	135

Each figure is the mean ± S.E. of the mean of six observations. The effect of the addition of glucose is in no case significant. The effect of insulin is significant in each instance ($P < 0·01$).

* These figures cannot be compared, as the experiments were performed at different times with different batches of ^{14}C-glycine in which the content of radioactivity had not been rigidly standardized.

acid incorporation and protein synthesis remain obscure.

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[1] Sinex, F. M., MacMullen, J., and Hastings, A. B., *J. Biol. Chem.*, **198**, 615 (1952).
[2] Krahl, M. E., *J. Biol. Chem.*, **200**, 99 (1953).
[3] Manchester, K. L., and Young, F. G., *Biochem. J.*, **70**, 353 (1958).
[4] Wool, I. G., and Krahl, M. E., *Amer. J. Physiol.*, **196**, 961 (1959).
[5] Wool, I. G., and Krahl, M. E., *Amer. J. Physiol.*, **197**, 367 (1959).
[6] Bruce, H. M., and Parkes, A. S., *J. Hyg.* (Camb.), **47**, 202 (1949).
[7] Gey, G. O., and Gey, M. K., *Amer. J. Cancer*, **27**, 45 (1936).
[8] Popják, G., *Biochem. J.*, **46**, 560 (1950).
[9] Manchester, K. L., and Young, F. G., *J. Endocrinol.*, **18**, 381 (1959).
[10] Kipnis, D. M., and Noall, M. W., *Biochim. Biophys. Acta*, **28**, 226 (1958).
[11] Manchester, K. L., and Young, F. G., *Biochem. J.* (in the press).
[12] Wool, I. G., and Krahl, M. E., *Nature*, **183**, 1399 (1959).
[13] Manchester, K. L., and Krahl, M. E., *J. Biol. Chem.*, **234**, 2938 (1959).

A HEPATOTOXIN CAUSING LIVER DAMAGE IN FACIAL ECZEMA OF SHEEP

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FACIAL eczema is a serious disease affecting sheep and cattle in many areas in New Zealand. The disease is essentially an acute cholangitis which, in more severe cases, is accompanied by obliteration of medium-size bile ducts, with bile ductule proliferation, and a gradual replacement of liver parenchyma in the portal areas with fibrous tissue as the lesions become more extensive[1]. It has recently been shown that this condition is due to a toxic principle present in the fungus *Sporidesmium bakeri*, Syd., a saprophytic organism growing in association with pasture[2]. Rabbits and guinea pigs are also known to be susceptible to the disease, liver lesions of a generally similar nature having been demonstrated in these species after feeding toxic grass and pure fungal culture[3,4], and ether extracts of these[4], and also after the injection of extracts into the vena porta[5].

Little is known of the behaviour of the toxin in the animal body. It has been generally assumed that the toxin is transported via the portal circulation to the liver and excreted into the biliary system, where it sets up an inflammatory reaction which in turn produces lesions characteristic of the disease[6]. There is, however, no experimental evidence to substantiate that the portal circulation is the main route by which the toxin enters the liver, nor is there any direct proof of the presence of the toxin or of a possible metabolite in the bile. These problems have recently been studied in some detail at this Station.

Transport of Toxin to the Liver

In an attempt to confirm the portal route of entry into the liver, and in order to obtain information on the rate of uptake of the toxin from the circulation, a comparison of the effectiveness of fungal extracts administered by the portal vein and by the jugular vein was made. A range of doses was tested on New Zealand white rabbits of mixed sex weighing 1,000–1,200 gm. Twelve animals were allotted to each group at random. All animals were slaughtered eight days after treatment. Morbid anatomy and histological examinations of specimens taken *post mortem* were carried out by D. C. Dodd, veterinary diagnostic officer, of this Station.

It has been found that over wide levels of dosage (ranging from the equivalent of 300 ml. to 1,000 ml. of fungal culture), macroscopic lesions of varying severity, consistent with facial eczema liver damage, were produced after intraportal injection of toxic extracts (Figs. 1, 2). The extent and severity of these

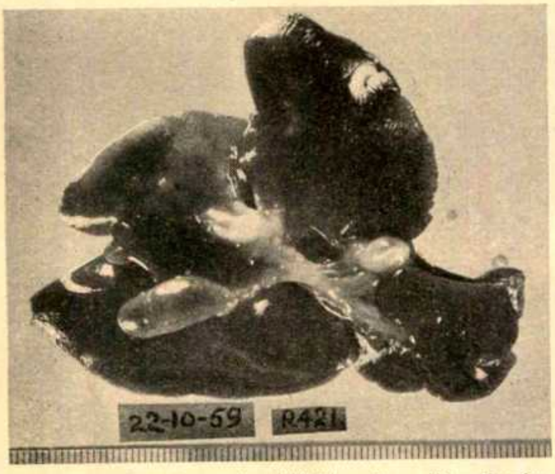

Fig. 1. Severe liver damage in rabbit fed a culture of *Sporidesmium bakeri*, Syd. for 14 days. Note extreme œdema of hepatic ducts, common bile duct and cystic duct, and marked fibrosis of left central lobe. Scale in millimetres

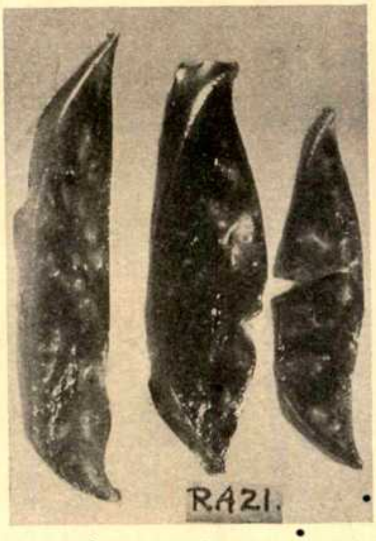

Fig. 2. Transverse sections of liver from same rabbit illustrating generalized damage to duct system

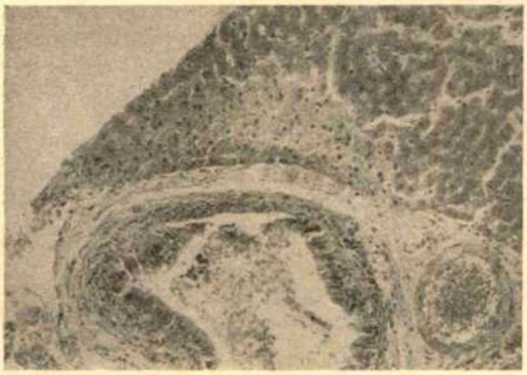

Fig. 3. Photomicrograph of local necrosis and desquamation of biliary epithelium of large bile duct with moderate periductal œdema and fibrosis. Hæmatoxylin and eosin. (×70)

lesions were confirmed histologically (Figs. 3, 4). After injection of comparable amounts of toxic material into the jugular vein, on the other hand, no evidence of any gross liver damage was found, and all livers, with the exception of two, were histologically normal. The two positive livers were graded as 'mild' on histological examination.

These observations provide support for the hypothesis that the portal circulation is the main route by which toxin enters the liver following absorption from the intestinal tract. The evidence also indicates that the toxin is relatively unstable in the systemic circulation and, on the basis of these findings, it would appear that uptake of toxin via the lymphatic system is of little or no importance in so far as the development of liver lesions is concerned. The fact that the toxin appears to be inactivated on entering the general circulation would suggest also that the uptake of toxin by the liver cells is instantaneous and that the lesions produced must, therefore, be due to that fraction of the toxin removed from the portal blood the first time it passes through the liver sinusoids.

Appearance of Toxin in the Bile

Prior to the work reported here, it had already been shown, in independent studies, that the first bile duct lesions begin to appear approximately three to six hours after intraportal injection of toxic extract[5], which strongly suggested that if toxin is excreted into the bile it must reach relatively high concentrations there very shortly after injection. It was decided, therefore, to collect bile from animals for varying intervals following injection of toxic extract and to test this bile directly for toxicity. Rabbits, of mixed sex, weighing 600–1,000 gm. were used. 'Donor' animals were fitted with a cannula into the common bile duct and injected intraportally with toxic extract equivalent to 5 litres of fungal culture. Toxicity was assessed by dripping the bile from the donor into the top of the duodenum of successive 'recipient' animals, up to six recipient animals being used in each experiment. This technique was decided upon after it had been shown independently that toxic extracts dispersed in fresh hepatic bile and administered in this way were effective in producing typical lesions of severe facial eczema liver. The technique offered the further advantage of minimizing the time for which any toxin present would be in contact with bile and also avoided the necessity for any form of chemical or physical treatment of the bile, which would have been necessary in order to reduce its bulk were it to be administered by any other route.

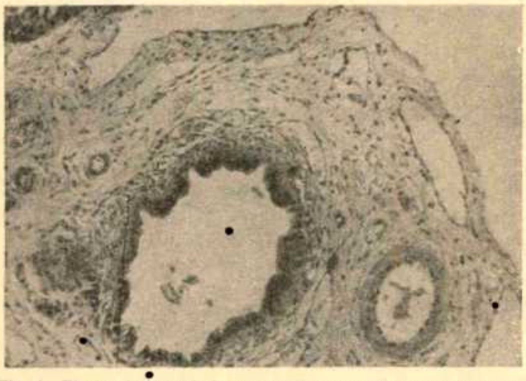

Fig. 4. Photomicrograph demonstrating typical severe periductal œdema and fibrosis, and cellular infiltration of periductal tissue. Hæmatoxylin and eosin. (×70)

Fourteen such experiments have been carried out to date. The technique employed in each has been essentially the same, with the exception that the intervals over which individual recipient animals received bile from the donor animal were varied in the different experiments from half an hour up to one and a half hours, although within each experiment the intervals were kept constant. In eleven of the fourteen experiments the presence of toxic material in the bile of the donor animal has been unequivocally demonstrated by the development of typical liver lesions of facial eczema in the recipient animals. The negative results obtained in three experiments were believed to be due to loss of toxicity of fungal extract prior to injection into the donor. In view of the wide individual variation in the degree of liver damage between animals which was observed in each experiment, it is difficult to assess with any degree of accuracy the rate of excretion of toxin from the liver. The evidence obtained so far does clearly indicate, however, that appreciable amounts of toxic material are present in the bile in as little as half an hour after injection of extract, and that maximal output appears to occur within two hours of injection. Whether the observed toxicity of bile was due to the presence of the toxin *per se* or to physiologically active degradation products of the toxin is not known. Attempts to settle this important issue must await elucidation of the chemical structure of the toxin.

The fact that toxicity has been detected in bile, using the technique outlined, is of considerable interest for our understanding of the pathogenesis of this disease, since it quite clearly provides strong evidence for supposing that in the intact animal an enterohepatic circulation of toxic material may occur. This might assist in explaining, at least in part, the 'progressive' nature of the disease[1], and the fact that such seemingly minute traces of toxin are capable of producing, in many cases, lesions of the most severe form (White, E. P., unpublished work).

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[1] Evans, J. V., McFarlane, D., Reid, C. S. W., and Perrin, D. D., *N.Z. J. Sci. Tech.*, **38**A, 491 (1957). Dodd, D. C., *Proc. N.Z. Soc. Anim. Prod.*, **19**, 48 (1959).

[2] Thornton, R. H., and Percival, J. C., *Nature*, **183**, 63 (1959).

[3] Percival, J. C., *N.Z. J. Agric. Res.* (in the press).

[4] Clare, N. T., *N.Z. J. Agric. Res.* (in the press).

[5] Worker, N. A., *N.Z. J. Agric. Res.* (in the press).

[6] McFarlane, D., Evans, J. V., and Reid, C. S. W., *N.Z. J. Agric. Res.*, **2**, 194 (1959).

LETTERS TO THE EDITORS

PHYSICS

Copper-Cellulose Complexes

THE criticism recently levelled by Bayley and Rose[1] at our findings concerning a copper–cellulose complex in wood implies that they have neither appreciated the weight of our evidence nor have they realized the complexity of the problems involved. We recommend that they should see all our papers on this matter[2-6]. We are answering these criticisms on behalf of the authors involved in our papers as we were responsible for the physical evidence presented.

When copper is taken up by wood and complexes in such a way as to be detectable by our methods, the question is whether the particular complex concerned is associated with the cellulose on one hand (as we believe we have proved), or with the encrusting substances (as Bayley and Rose would have us believe) on the other. This latter possibility must be rejected on several clear grounds. (*a*) The complex gives identically the same electron diffraction patterns whether whole wood or wood α-cellulose is used and, moreover, these are invariable for a wide range of α-celluloses and a wide range of metals. (*b*) The complex is oriented with respect to the cellulose microfibrils. This is demonstrated both by the electron diffraction diagram and by the dichroic properties resulting when wood or α-cellulose is treated with a copper salt and stained with dithioxamide. There is no evidence that the encrusting substances have the requisite properties. (*c*) The uptake/concentration curves demand that the copper shall be adsorbed on a surface, and the electron diffraction diagrams prove that the copper ions then form a regular two-dimensional array. The microfibrils provide such a regular surface, but there is no evidence that the encrusting substances fulfil this requirement.

We have emphasized in our papers that copper, in particular, occurs in more than one form in wood impregnated with copper salts and of course it may form a complex with some of the encrusting materials. We have been careful not to exclude this possibility though we have at present no evidence for it; our evidence refers exclusively to a copper–microfibril complex. It is not quite certain that the complex involves any part of the glucose residue in cellulose, and non-glucose sugars that are known[7] to be an integral part of the cellulose microfibril may be involved. The facts, however, that the same electron diffraction diagram is obtained for celluloses varying widely in xylose content, and that if non-glucose sugars are involved they would need to be closely and regularly distributed over the surface of microfibrils, argue strongly in favour of glucose. For the same reasons we are of the opinion that the hydroxyl groups of glucose are involved rather than occasional carboxyl substituents.

It is perhaps at first sight peculiar that microfibrils embedded in lignin and hemicelluloses possess the necessary free surfaces, and it is presumably this that formed the starting point of Bayley and Rose's[1] comments. It must be remembered, however, that by no means all of the microfibrils either in wood or in wood α-cellulose become complexed. The evidence so far is that less than about a quarter of the total microfibrillar surface is apparently capable of adsorbing metals in the manner under consideration. The evidence shows that this much of the surface is available in wood, and unless our evidence can be shown to be at fault this must be accepted.

We would suggest that, until firm evidence to the contrary has been presented, our hypothesis is the only reasonable interpretation of the facts.

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[1] Bayley, C. H., and Rose, G. R. F., *Nature*, **185**, 313 (1960).

[2] Belford, D. S., Preston, R. D., Cook, C. D., and Nevard, E. H., *Nature*, **180**, 1081 (1957).

[3] Belford, D. S., Myers, A., and Preston, R. D., *Nature*, **181**, 1516 (1958).

[4] Belford, D. S., Preston, R. D., Cook, C. D., and Nevard, E. H., *J. App. Chem.*, **9**, 192 (1959).

[5] Belford, D. S., Myers, A., and Preston, R. D., *Biochim. Biophys. Acta*, **34**, 47 (1959).

[6] Belford, D. S., Cook, C. D., Nevard, E. H., and Preston, R. D., *Nature*, **183**, 988 (1959).

[7] Cronshaw, J., Myers, A., and Preston, R. D., *Biochim. Biophys. Acta*, **27**, 89 (1957).

Sorption of Activated Gases by Titanium Films

GALE[1] has shown that the rate of gas sorption of an evaporated titanium film is enhanced by a Penning gauge connected to the getter vessel, and experiments are in progress in my laboratory to determine the nature of the sorption process when the gas is activated in this manner. In a recent communication it has been shown that the rate of gas sorption of titanium films for air, nitrogen, oxygen and hydrogen are a function of the current flowing in the Penning head[2], even when the metal film is at a positive potential and therefore repels positive ions. The increase in the rate of sorption of active gases greatly exceeds that due to the pumping action of the Penning head, but inert gas atoms are only sorbed by the Penning head and are probably embedded in metal sputtered from the cold cathodes.

I have proposed[3] that the enhanced rate of sorption of an active gas arises primarily from the Penning discharge atomizing the gas, so that it diffuses more readily between the grain boundaries of the metal deposit. Possibly the active states of the gas particles also increase their rate of chemical reaction at the nascent metal surface; but this can scarcely explain an enhanced rate of sorption if one considers that sorption proceeds through a superficial layer of reaction products. Titanium films evaporated at low pressure (< 0·1μ mercury) have almost bulk density, because when oxidized by heat the resulting layer of titanium oxide has a refractive index near that of solid rutile in the polycrystalline state. Evaporated titanium films grow from a large number of nuclei, and the condensate must be composed of tightly packed micro-crystallites possessing a large interior area for adsorption and reaction with residual gases. However, the rate of sorption of a fresh film would rapidly decline, because the gas molecules are unable to reach the interior surface. Dependence of the rate of diffusion of the sorbed gas on the structural form of the deposit may account for the conflicting reports of the effects of ionization (or atomization) of gases on the rate of sorption of barium (see, for example, refs. 3 and 4), because with this metal the grain size and distribution can vary considerably with the evaporation conditions.

The investigations of the gettering properties of titanium have recently been refined and show more clearly the dependence of the rate of sorption on the ionization (atomization) of the residual gas. The Penning ion-source was mounted in a 3-in. diameter tube with titanium cathodes of 3 in. diameter and in a magnetic field-strength of 400 gauss. When operated at applied voltages between 3 and 5 kV., the ion-source had a pumping speed of about 1–5 litre/sec. The sorption vessel consisted of a stainless steel chamber 6 in. in diameter and 6 in. high (1,400 cm.^{2} area) with the Penning head mounted on top. The positive terminal of the high-tension supply was grounded, so that the evaporated getter repelled gas ions. The vacuum seals were made with metal gaskets and the pressure measured with an Alpert type ion-gauge. The titanium was evaporated from tungsten filaments after baking the system and isolating the chamber from the oil diffusion pump at a pressure of 10^{-7} mm. mercury. The gas to be sorbed was admitted to the getter chamber from a reservoir through a calibrated leak. Rates of sorption were measured in the pressure interval 10^{-7} to 10^{-5} mm. mercury.

In the earlier work[1], a thick titanium film was used for a series of sorption measurements over a range of pressures. In this investigation a fresh film (≃ 100 Å. thick) was used for each sorption measurement, and a period of 1 min. was allowed after each evaporation for the system to gain equilibrium before the instantaneous rate of sorption was measured. At the pressures used, the outer surface of the getter film was rapidly covered by adsorbed gas, and this was shown by the fact that the rate of sorption fell to a negligible amount ($< 0{\cdot}002$ litre μ/sec.) after a few minutes if the Penning head was not on. In Fig. 1 are shown the mass sorption-rates of titanium films for oxygen when measured at different pressures and under the foregoing conditions. The Penning head was operated at constant voltages of 3 and 5 kV., and the current flowing in the head has been plotted in Fig. 1 as a function of pressure. When the two sets of curves are compared, it can be seen that the rate of sorption is practically a linear function of the Penning discharge current, which itself is not exactly proportional to pressure. At high values of ionization current the rate of sorption tends to increase less rapidly, presumably because the getter film is more quickly saturated by the gas.

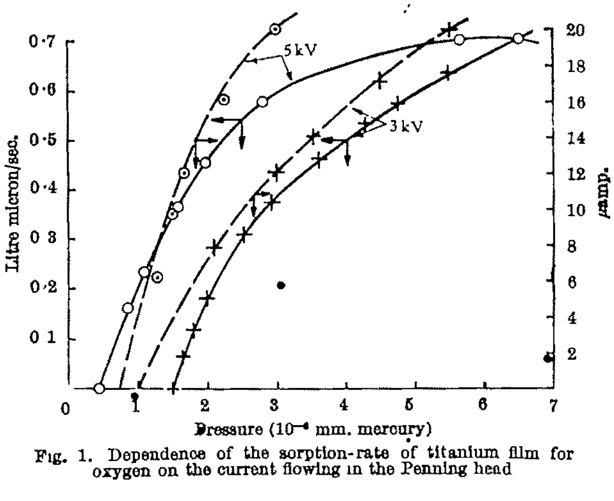

Fig. 1. Dependence of the sorption-rate of titanium film for oxygen on the current flowing in the Penning head

In a recent communication in *Nature*, Stow[5] reported that titanium films deposited on the negative ion collector of a hot-cathode ion-pump gave the same rate of sorption whether the ion-pump was in operation or not. From this he concluded that ionization did not play an essential part in the sorption process. However, his pressure measurements were made with a Penning gauge and if, as suggested here, such a discharge supplies atomized neutral particles to the film, then operation of the gauge would have enhanced the rate of sorption in both cases. The ionization (and related dissociation) efficiency of a Penning system is known to be superior to that of a triode type ion-source. These criticisms do not in any way detract from his substantiation of the value of titanium as a getter. Stow's subsequent experiments comparing barium and titanium getters were based on measurements made with Alpert ion-gauges, and molecular dissociation could have still been effective.

The theory advanced in this communication to explain the enhanced rate of sorption of a deposit also accounts for the enhanced rate of oxidation of thin metal films in glow discharges. There is no evidence that oxide films on metals grow faster in the presence of active oxygen, but penetration of a metal film by gas atoms so that the crystallites are oxidized on many sides would explain this phenomenon.

Acknowledgments are made to Mr. W. Sheraton, who made the sorption measurements described here, and to the Directors of Edwards High Vacuum, Ltd., for permission to publish this communication.

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[1] Gale, A. J., C.V.T. Vacuum Symposium Trans., 12 (Pergamon Press, London, 1956).
[2] Holland, L., and Harte, A., Int. Symp. Residual Gases in Electron Tubes and Related High Vacuum Systems, Como, Sept. 1959 (Pergamon Press, London, to be published).
[3] Wagener, S., *Nature*, 173, 684 (1954).
[4] Cloud, R. W., Beckman, L., and Trump, J. G., *Rev. Sci. Instr.*, 28, 889 (1957).
[5] Stow, R. L., *Nature*, 184, 542 (1959).

Sedimentation and Effective Viscosity

I WISH to comment on the communication by Prof. G. J. Kynch on this subject[1]. With the author's reservations (see following letter), the derivation of equation (8) seems satisfactory, though it is difficult to see the need for the constant added in equation (6). The final equation may be written in the form :

$$V_r = \frac{u}{V} = \frac{(1 - c)}{\mu_{re}} \tag{1}$$

where V_r is relative settling velocity of particles; u is actual settling velocity of particles; V is settling velocity of a single typical particle at infinite dilution;

c is volume concentration of solids; μ_{re} is effective relative viscosity of suspension of similar particles having the same density as the fluid and a volume concentration of c. This equation makes interesting comparison with equations due to Gurel[2],

$$V_r = \frac{(1-c)^2}{\mu_r} \qquad (2)$$

where μ_r is the measured relative viscosity of the suspension, and Hawksley[3],

$$V_r = (1-c)^2 \exp - \frac{2\cdot5c}{1-0\cdot609c} \qquad (3)$$

and Steinour[4],

$$V_r = (1-c)^2\, 10^{-1\cdot82c} \qquad (4)$$

All four equations contain a term which represents the reciprocal of the effective relative viscosity of the suspension; this is multiplied by $(1-c)^2$ in equations (2), (3) and (4), and by $(1-c)$ in equation (1). The latter term is due to the familiar 'return flow' effect, also considered by the other workers; the additional factor $(1-c)$ in equations (2), (3) and (4) appears because a typical particle is considered as settling in a fluid of density equal to that of the suspension as a whole rather than one of density equal to that of the dispersing liquid. Kynch, however, considers a typical particle differing from its fellows only in that its density is equal to that of the dispersing liquid. The time-average velocity of this particle is stated to equal the mean fluid velocity "modified by the pressure gradient in the fluid due to the [other] falling particles". Following the application of an equation in which the density of the particle reverts to its original value, a value of the settling velocity is obtained by neglecting the above pressure gradient. Equation (1) is therefore advanced only as an 'approximate' relationship for the settling velocity.

It is then stated that the equation "seems to agree with experimental results up to concentrations of about 20 per cent", ref. 5 being quoted. This agreement is actually rather poor, for the curve referred to shows that the product $\mu_r V_r$ is proportional to $(1-c)^2$ rather than $(1-c)$; thus distinct non-linearity is present at concentrations of less than 20 per cent when the data are plotted on the basis of $\mu_r V_r$ against $(1-c)$. Moreover, in the derivation of equation (1), it was stated that the viscosity was that of an otherwise similar suspension in which the particles had the same density as the fluid, that is, a stable suspension. Accordingly, a graph has been drawn in which the relative viscosities used are for stable suspensions of spherical particles[6] and the settling velocities for otherwise similar slightly unstable suspensions having particle size ratios of 1·40 : 1 (ref. 7) and 1·03 : 1 (ref. 2), respectively (Fig. 1). In neither case does equation (1) apply satisfactorily, although for the suspensions having particles of wider size-range the gradient of the line near the origin is not greatly different from that predicted by the theory. The suspensions of closely sized particles represent an important limiting case on which few data are available; the unique settling behaviour of these suspensions merits further attention.

It should be added in fairness that equation (2) also requires modification before agreement with experimental results is obtained[8]. Fair agreement with the present data has, however, been given by equation (3), linear plots being obtainable for concentrations up to more than 30 per cent with gradients of 1·26 (size ratio 1·03 : 1) and 0·88 (size ratio 1·40 : 1), compared with a theoretical value of 1·09. These figures illustrate the importance of reducing the size-range of particles used in sedimentation tests if the results are to be compared with elementary theory.

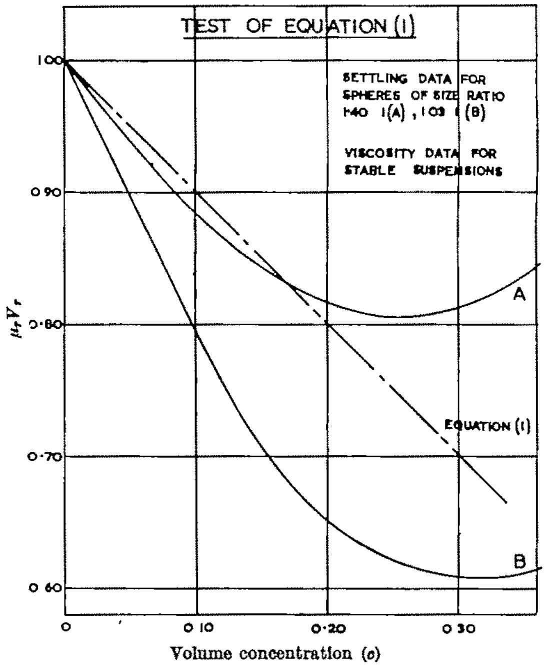

Fig. 1

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[1] Kynch, G. J., *Nature*, 184, 1311 (1959).

[2] Gurel, S., Ph.D. thesis (Birmingham, 1951).

[3] Hawksley, P. G. W., "Some Aspects of Fluid Flow", 114 (Inst. Phys., 1951).

[4] Steinour, H., *Indust. Eng. Chem.*, 36, 618, 840 (1944).

[5] Ward, S. G., *J. Oil and Colour Chem. Assoc.*, 38 (1955).

[6] Oliver, D. R., and Ward, S. G., *Nature*, 171, 396 (1953).

[7] Oliver, D. R., Ph.D. thesis (Birmingham, 1954); see Table L, Class 2, Col. 2).

Before I comment on Dr. Oliver's communication, I feel that an apology is due to readers for a number of errors in my communication in *Nature* of October 24. They made a simple derivation of equation (8) appear complicated though not, I hope, unintelligible.

Dr. Oliver's comments are valuable, as he is familiar with the difficulties of this subject, and I am glad that he considers that the derivation of my equation seems satisfactory and produces no theoretical reasons for disagreeing with it. Incidentally, the other equations which he mentions are referred to and commented upon by Ward in the reference given in my letter.

At present the experimental results on suspensions seem complicated and often have surprising features. Objections can be raised to all theoretical formulæ or semi-empirical formulæ yet suggested, and the reasons for the complications are not fully understood. In these circumstances, as in many others, the most useful theory is simple, generally acceptable, containing well-defined approximations with some physical meaning. It may then be possible to suggest reasons for discrepancies between the approximate theory and accurate experiment which will lead to an improved formula. I hope that the derivation in my letter might help in this way and suggest further experiments.

Dr. Oliver clearly does not accept my statement that my equation "seems to agree with experimental results up to concentrations of 20 per cent", although he admits that the other equations are not good either. He mentions my reference to the article by Ward, where there is certainly a curve intended to show that $\mu_r V_r$ is proportional to $(1 - c)^2$ rather than $(1 - c)$. I was not entirely satisfied with this conclusion and I looked at the data used in the article, which were obtained by Oliver[2]. In fact, his results for 'Kallodoc' spheres in water and in an alcohol–water mixture and for glass spheres in glycerol–water agree quite well with my formula, as I stated, and at least as well as with the formula with the factor $(1 - c)^2$. Only for 'Kallodoc' spheres in lead-nitrate solution was there serious error and for suspensions with nearly equi-sized particles. Dr. Oliver plots two curves using viscosity data for stable suspensions, one giving fair agreement and the other, for nearly equi-sized particles, giving bad agreement.

From my equations (1)–(4), which I reproduce in order:

$$v = v_f + v_p : \quad U = v' - v : \quad U\mu = V\mu_0 : \quad v' = u$$

together with the assumption $v_f = -cu/(1 - c)$, I deduce that:

$$\mu u = (1 - c)(\mu_0 V + \mu v_p)$$

The discrepancies in my simple formula can be attributed to its assumption that $v_p = 0$, or to the other assumption that μ is the effective viscosity μ_{re}.

Whatever the cause, the discrepancy for equi-sized spheres is puzzling and requires more consideration.

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[1] Ward, S. G., *J. Oil and Colour Chem. Assoc.*, 38 (1955).

[2] Oliver, D. R., Ph.D. thesis (Birmingham, 1954).

METALLURGY

Influence of Guinier–Preston Zones on the Electrical Resistivity of Age-hardening Alloys

Federighi and Passari[1] have recently directed attention to the increase in electrical resistivity associated with the early stages of ageing in binary aluminium alloys in which age-hardening involves pre-precipitation effects such as the formation of Guinier–Preston zones. Recent work in this Establishment has shown that this effect occurs in more complex ternary and quaternary aluminium alloys and is particularly marked in the Al–Zn–Mg–Cu system.

The effect is shown in Fig. 1, which compares present observations on an Al–Zn–Mg alloy with earlier work by Durer and Köster[2]. Ageing at 20° C. yields a significant increase in resistivity whereas at high temperatures the normal fall in resistivity associated with precipitation is observed. Very similar results have now been obtained on commercial Al–Mg–Si alloys (Fig. 2) and the effect appears to be typical of a number of binary and ternary aluminium alloys, including duralumin type alloys[3], all of which are known to develop Guinier–Preston zones in the initial stages of hardening and particularly during low-temperature ageing. The lack of coincidence between the resistivity and hardness increases has been ascribed to the dependence of the resistivity on electron scattering by aggregates of criticial dimensions and on the effect of coherency strains in the matrix[4,5]. In the Al–Zn–Mg system it is known that low-temperature ageing involves the segregation of Zn–Mg zones coherent with the matrix which develop to dimensions of the order of 25 Å. × 50 Å. diameter, followed by the development of an intermediate, hexagonal precipitate which gradually loses coherency with the matrix. However, Guinier–Preston zone formation undoubtedly occurs in the early stages of high-temperature ageing as well, for such zones are readily developed during quenching and heating to the ageing temperature. If the above hypothesis is correct then the loss of resistivity in the early stages of high-temperature ageing must be ascribed to accelerated growth of the zones beyond critical dimensions and earlier development of intermediate precipitates which are not markedly coherent with the matrix.

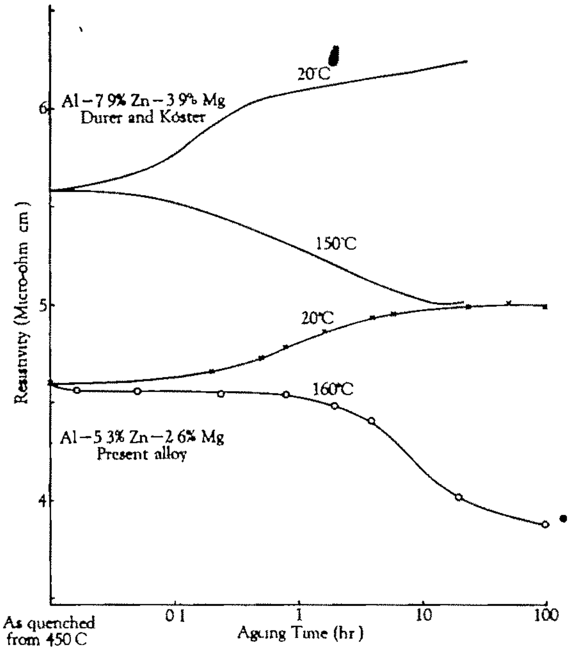

Fig. 1. Isothermal ageing Al–Zn–Mg alloys

Fig. 3 shows the profound effect of adding copper to an Al–Zn–Mg alloy of similar composition to that in Fig. 1. The ageing process is retarded and the resistivity changes are far more marked and persist to higher ageing temperatures. Single-crystal X-ray work on these alloys has shown that the Guinier–Preston

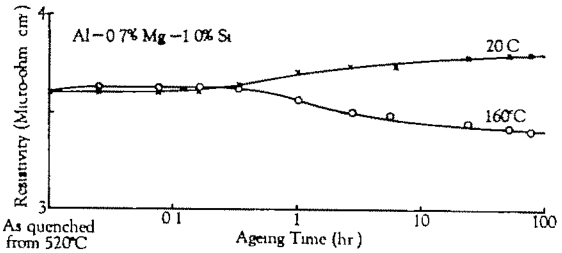

Fig. 2. Isothermal ageing Al–Mg–Si alloy

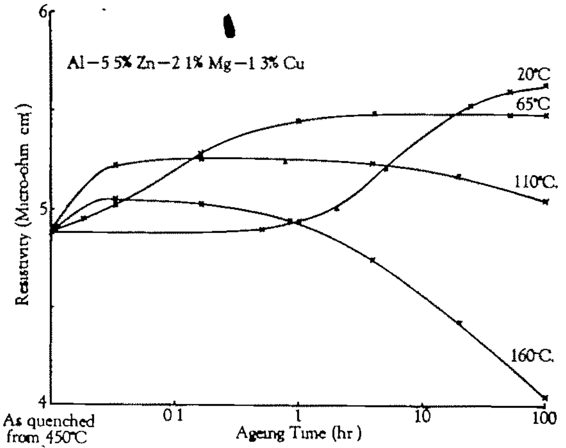

Fig. 8. Isothermal ageing Al–Zn–Mg–Cu alloy

zone structure once established is particularly stable and persists even during ageing at 160° C. and higher. The experimental evidence suggests that ideally the zone structure would not develop at ageing temperatures above 110° C., but in practice does so during heating to the ageing temperature (heating time less than 1 min.). For ageing temperatures of 110° C. and above the resistivity changes and mechanical properties of this alloy are significantly affected by the rate of heating to temperature, both peak hardness and peak resistivity being increased by slow heating.

For these higher ageing temperatures the resistivity peak is closely associated with the primary hardness increase, and it is reasonable to suppose that the resistivity effect is more closely associated with high coherency strains rather than with the existence of zones of critical dimensions. Thomas and Nutting[6] estimate that the zone diameter in this quaternary material is of the order of 80 Å.

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[1] Federighi, T., and Passari, L., *Acta Met.*, 7, 422 (1959).

[2] Durer, A., and Koster, W., *Z. Metall.*, 30, 311 (1938).

[3] Cohen, M., AIMME Metals Technology Tech. Paper 978 (1938).

[4] Mott, N. F., *J. Inst. Met.*, 60, 267 (1937).

[5] Geisler, A. H., Symp. Phase Transformations in Solids, 387 (Wiley, New York, 1951).

[6] Thomas, G., and Nutting, J., *J. Inst. Met.*, 88, (2), 81 (1959).

Purification of Metals by Gas Chromatography

The use of chromatographic techniques for the separation of an alloy into its constituent elements or for the direct purification of metallic elements has not, to our knowledge, been reported. In order to test the feasibility of the separation a zinc–cadmium (8·4 per cent zinc) alloy was used. The advantages of this alloy system for this test are: (1) both elements have significant vapour pressures at temperatures which do not pose extremely difficult material problems; (2) they are chemically similar; (3) they are common metals; (4) their reactivity is small enough so that they pose no handling problems (as would the alkali metals).

The containing tube was a 50-cm. length of 13-mm. 'Vycor'. The stationary phase, lithium chloride, was supported as a film on sea sand (20 per cent lithium chloride, 80 per cent fine sea sand). The temperature was high enough so that the lithium chloride melted and formed a film on the sand. The eluting gas, purified helium, had a velocity of 150 ml./min. and was preheated to 620° C., the bed temperature. The 0·1-gm. sample of alloy was laid on top of the bed before heating. It was necessary to eliminate most of the oxygen and water from the system in order to minimize oxidation of the sample. The emerging metal vapours were caught on glass cold fingers held in the gas stream for a short time. The metal was dissolved in distilled nitric acid and analysed spectrographically.

It was shown that the cadmium content of the vapour was more than 200 times the zinc content. The amount of zinc present was near the lower limit of the analytical procedure. It was also shown, by duplicating the above experiment without the stationary or solid phases, that the process was not that of simple distillation. In this case the zinc deposit was equal to, or greater than, the cadmium deposit. Although neither of the constituents was collected quantitatively, these experiments suggest that metals can be efficiently separated.

I wish to acknowledge the assistance of Messrs. S. Greenberg and C. A. Youngdahl. This work was done under the auspices of the U.S. Atomic Energy Commission.

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CHEMISTRY

Decomposition of Uranium Dioxide at its Melting Point

We have found that fusion of uranium dioxide in an argon arc furnace at 2,800 ± 100° C., between a tungsten cathode and a water-cooled copper anode, consistently gave lustrous grey metallic-looking products, with the gross analytical composition UO_{2-x} $(0 \cdot 15 > x > 0)$. The fused material was contaminated with less than 0·001 per cent of copper or tungsten. The extent to which the product was deficient in oxygen depended upon how long the sample had been held in the molten state, but the liquid seemed to approach a limiting composition, about $UO_{1 \cdot 88}$, after prolonged fusion. There was some vaporization from the melt, but the constitution of the vapour phase is unknown; after exposure to air, the powdery sublimate present in the furnace consisted of the oxygen-excess phase UO_{2+x}.

The massive fused material was quite stable in air, but when it was powdered it readily combined with oxygen, up to a composition around $UO_{2 \cdot 005}$. X-ray analysis of a fine chip of the fused product revealed the presence of two phases.

(*A*) The major phase (estimated 94–98 mole per cent) was face centred cubic with $a_0 = 5 \cdot 4718 \pm 0 \cdot 0004$ Å., which may be compared with the fluorite cell of essentially stoichiometric uranium dioxide, $UO_{1 \cdot 999 \pm 0 \cdot 001}$, with $a_0 = 5 \cdot 4700 \pm 0 \cdot 0004$ Å.

This small but significant expansion of the uranium dioxide lattice, which has also been observed when uranium metal and uranium dioxide are heated above 2,000° C. in a graphite resistance furnace[1], is taken to

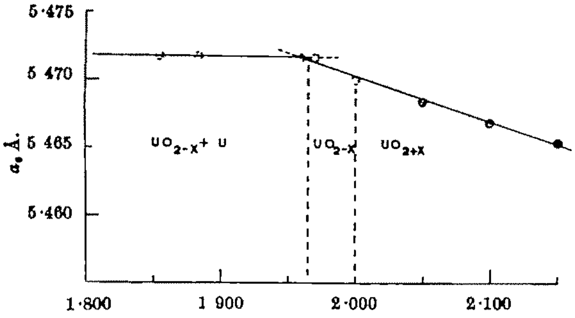

Fig. 1. ○, Ackermann's values

indicate that uranium metal is slightly soluble in uranium dioxide, that is, that the stoichiometric range of uranium dioxide extends slightly on the oxygen-deficient side of the ideal formula, at the quenching temperature of the arc furnace (estimated as between 2,500° C. and 1,600° C.). Extrapolating from the recent data of Ackermann[2] for the cell dimensions of oxygen-excess uranium oxides, it can be deduced that the limiting composition of the oxygen-deficient phase is $UO_{1\cdot 96}$ to $UO_{1\cdot 97}$ (Fig. 1).

(*B*) A minor phase was present to the estimated extent of 2–6 mole per cent. Its diffraction pattern consisted of seventeen lines, the *d* values of which are irreconcilable with the presence of any of the following phases: (*a*) the reported cubic lower oxides—uranium monoxide[3] with $a_0 = 4 \cdot 92$ Å., Zachariasen's supposed $UO_{1\cdot 75}$ (ref. 3) with $a_0 = 5 \cdot 477$ Å., or the UO_{2-x} of Perio and Hering[4] with $a_0 = 5 \cdot 484$ Å; or (*b*) the α- or γ-allotropes of uranium metal. α-Uranium, stable between room temperature and 667° C., is orthorhombic with *a* 2·852 *b* 5·865 *c* 5·495 Å. (refs. 2, 5); γ-uranium, stable above 767° C., is body centred cubic, $a_0 = 3 \cdot 43$ Å. (refs. 2, 3). However, the interplanar spacings agree fairly well with those of lines in the complex diffraction pattern[6] of β-uranium, the form stable only between 667° C. and 767° C. (Table 1).

Table 1. COMPARISON OF X-RAY DATA FOR THE MINOR PHASE WITH THE X-RAY DATA FOR β-URANIUM

β-Uranium (6)		Minor phase		β-Uranium		Minor phase	
d Obs.	*I* Obs.	*d* Obs.	*I* Obs.	*d* Obs.	*I* Obs.	*d* Obs.	*I* Obs.
2·827	10	2·77	w.	1·163	20	1·16	w.
2·696	4			1·116	18		
2·649	6			1·079	5		
2·612	70	2 58	w.	1 073	9		
2 536	53			1 051	24		
2 500	41	2·50	m.	1 037	22		
2·438	48			1 035	19		
2·370	100	2·39	m.	1 022	36	1 02	w.
2·313	58			1 005	15	1·01	w.
2·270	20	2·28	w.	0·9947	18		
2·175	19	2·17	m.	0·9796	25		
2 054	11			0·9742	24		
2 011	9			0 9621	20		
1 975	9	1 96	v.w.	0 9489	23	0 954	w.
1·712	11	1 71	w.				
1 692	14						
1 631	25						
1·615	10						
1·543	28	1 56	w.				
1 526	45						
1·513	40						
1·502	42						
1·479	37	1·48	w.				
1 467	19						
1 457	17						
1 442	21						
1·415	13						
1·304	24	1 30	w.				
1·289	6						
1·243	29						
1·214	22						
1 207	22	1·21	w.				
1·193	9						
1·184	17						
1·177	10	1 17	v.v.w.				

m, Medium; w., weak; v.w., very weak; v.v.w., very very weak.

Proof that the minor phase was uranium metal was afforded by the following experiment. One of the fused samples, of composition $UO_{1\cdot 883}$, was powdered and sealed into an X-ray capillary, both operations being carried out inside a sealed, evacuated apparatus. The X-ray pattern then showed that while the major phase was unchanged, the minor phase had altered; its diffraction pattern could be completely indexed in terms of the α-uranium structure (Table 2).

Table 2. COMPARISON OF X-RAY DATA FOR THE MINOR PHASE (AFTER POWDERING) WITH THE X-RAY DATA FOR α-URANIUM

α-Uranium (5)		Minor phase		α-Uranium		Minor phase	
d Obs.	*I* Obs.	*d* Obs.	*I* Obs.	*d* Obs.	*I* Obs.	*d* Obs.	*I* Obs.
2·563	s.	2 562	w.	1 012	m.	1·014	v.w.
2·521	s	2 521	w.	0 992	m.	0 993	v.w.
2·473	s.	2·475	w.	0 945	m.		
2·274	s.	2 275	w.	0·936	m.		
1·778	s.	1 781	w.	0 922	w.m.		
1·532	s.	1·533	v.w.	0 878	w m.		
1 436	m.	1·437	v.w.	0·848	s.	0·844	v.v.w.
1·387	m.			0·815	s.	0·816	v.v w.
1 260	m.						
1·238	s.						
1·153	m	1 153	v.w.				
1·114	m.						
1·088	w.m.						

s, Strong; m., medium; w., weak; v.w., very weak; v.v.w., very very weak.

Thus uranium dioxide decomposes at its melting point, first to an oxygen-deficient oxide and then to metallic uranium (Fig. 1). Under the conditions of our experiments the metal, dispersed through the fused oxide, appeared to be retained in the highly metastable β-modification, but reverted to the stable α-uranium when the sample was ground up in the absence of oxygen.

Further work on the uranium–uranium dioxide system at high temperatures is being carried out and will be described elsewhere.

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[1] Rundle, R. E., Baenziger, N. C., Wilson, A. S., and McDonald, R. A., *J. Amer. Chem. Soc.*, 70, 99 (1948).

[2] Ackermann, R. J., *AEC–ANL*–5482 (1955).

[3] Katz, J., and Rabinowitch, E., "The Chemistry of Uranium", Part 1 (New York, 1951).

[4] Hering, H., and Perio, P., *Bull. Soc. Chim. France*, 351 (1952).

[5] Jacob, C. W., and Warren, B. E., *J. Amer. Chem. Soc.*, 59, 2589 (1937).

[6] Thewlis, J., *Acta Cryst.*, 5, 790 (1952). Tucker, C. W., Senio, P., Thewlis, J., and Steeple, H., *Acta Cryst.*, 9, 472 (1956).

Electron Spin Resonance Absorption of the Biradical Molecular Complex, *p*-Phenylenediamine-chloranil

ACCORDING to Mulliken's quantum-mechanical theory the lowest state of a molecular complex *A.D*, where *A* and *D* stand for the electron acceptor and donor, respectively, is chiefly a non-bonding state, and the next excited state is a dative state $A^- – D^+$ (ref. 1). Kainer *et al.*[2–5] have considered the existence of molecular complexes where the ionic state is the lower of these two states and studied the electron spin resonance and the other properties of a number of

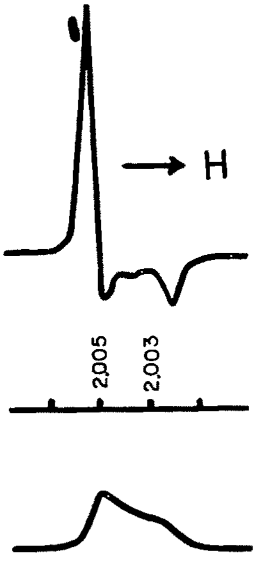

Fig. 1. Electron spin resonance absorption spectrum of the molecular complex *p*-phenylenediamine-chloranil

molecular complexes which consist of aromatic diamines and quinones.

It has been known that even in a biradical molecule of Chichibabin's hydrocarbon type, or its analogues, two unpaired electrons are only weakly coupled[6,7]. Therefore, it seemed that it might be possible to resolve the resonance absorption of aromatic diamine-halogenoquinone complexes because of a fairly large difference of *g*-value between these two components in ionized forms. With this hope we re-examined the electron spin resonance absorption of some aromatic diamine-halogenoquinone complexes using a high-resolution spectrometer[8]. The measurements were at a frequency of 9 kMc./s. and at room temperature.

Only in the case of *p*-phenylenediamine-chloranil was the resonance absorption clearly resolved and the two peaks were definitely correlated with the component ions. It was found that the shape of the resonance absorption curve depends, to some extent, on the preparation. A typical spectrum is shown in Fig. 1. The spectrum consists of a sharp peak with a *g*-value of 2·005 and a slightly broader one with 2·003. The former *g*-value is in good agreement with that of the potassium salt of tetrachloro-*p*-benzosemiquinone, 2 0048, and the latter with those of Wurster's blue salts, 2·0028 for the ferricyanide, and 2·0033 for the picrate[9]. The spectrum of N,N,N′,N′-tetramethyl-*p*-phenylenediamine-chloranil could not be well resolved but was found to be highly asymmetric. The main peak is again located at the position of tetrachloro-*p*-benzosemiquinone, but the slope at low magnetic field side is much steeper.

Our results seem to show that these molecular complexes, which we may call salts, consist of ionized components, and the electronic interaction between positively charged diamine and negatively charged quinone molecules is not strong. Both these molecular complexes are unstable. The colour of the latter sample changed from dark red to blue on standing for a week and its spin concentration decreased by a factor of ten. The same blue product could be obtained by mixing warm benzene solutions of the components. Tetrabromo-*p*-benzosemiquinone has a higher *g*-value than its chloro-analogue; however, the molecular compounds with bromanil gave absorptions which were too broad to be resolved.

We also detected a weak resonance absorption in *p,p*-diethoxybinaphthostilbene-chloranil[10]. It is not clear whether or not this molecular complex is of the ionic type like aromatic diamine-quinone.

The spin concentration in these aromatic diamine-quinone complexes has been found to be independent of temperature[3,5]. This property has not yet been well explained[5]. In connexion with this, it may be interesting to recall the observation that polycyclic aromatic hydrocarbon-iodine complexes, where the ground-state is supposed to be non-bonding, show strong electron spin resonance absorption[11]. No change of spin concentration in these molecular complexes was observed in the range from 77° K. to room temperature.

We wish to thank the National Research Council of Canada for grants in aid of this work.

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[1] Mulliken, R. S., *J. Amer. Chem. Soc.*, **74**, 811 (1952).

[2] Kainer, H., Bijl, D., and Rose-Innes, A. C., *Naturwiss.*, **41**, 303 (1954).

[3] Kainer, H., and Überle, A., *Chem. Ber.*, **88**, 1147 (1955).

[4] Kainer, H., and Otting, W., *Chem. Ber.*, **88**, 1921 (1955).

[5] Bijl, D., Kainer, H., and Rose-Innes, A. C., *J. Chem. Phys.*, **30**, 765 (1959).

[6] Jarrett, H. S., Sloan, G. J., and Vaughan, W. R., *J. Chem. Phys.*, **25**, 697 (1956).

[7] Reitz, D. C., and Weissman, S. I., *J. Chem. Phys.*, **27**, 968 (1957).

[8] Corfield, W., Farmer, J. B., Horsfield, A., and McDowell, C. A. (to be published).

[9] Holden, A. N., Yager, W. A., and Merritt, F. R., *J. Chem. Phys.*, **19**, 1319 (1951).

[10] Pfeiffer, P., *Ann.*, **404**, 1 (1914).

[11] Matsunaga, Y., *J. Chem. Phys.*, **30**, 855 (1959).

Spectrum of Delayed Fluorescence in Phenanthrene Vapour : a Criterion of Purity

In addition to the normal fluorescence emission with a decay constant of $\sim 10^8$ sec.$^{-1}$, a slow component with a decay constant of $\sim 10^3$ sec.$^{-1}$ has been detected by Dikun in the luminescence of phenanthrene vapour[1]. On the grounds that the spectra of both components are identical, Williams[2] has attributed this delay to the reversible formation of an excited dimer, from normal and electronically excited molecules, which has a life-time of the order of a millisecond.

In view of the remarkable similarity between the phenanthrene vapour emission spectra obtained by Dikun and the fluorescence spectrum of anthracene vapour, we decided to repeat this work using Eastman Kodak phenanthrene of 'highest purity, which was treated with maleic anhydride[3] until a saturated solution in hexane was completely transparent above 355 mμ ; this places an upper limit of one part in 25,000 on the concentration of anthracene impurity.

The phenanthrene vapour at 9·6 mm. and 260° C. was irradiated in a 'Pyrex' cell by the 313 mμ line isolated from a high-pressure mercury-vapour lamp by an alkaline chromate filter[4]. Emission spectra were photographed on a small Hilger quartz spectrograph, a locally constructed phosphoroscope being used to isolate the slow component ; a medium Hilger

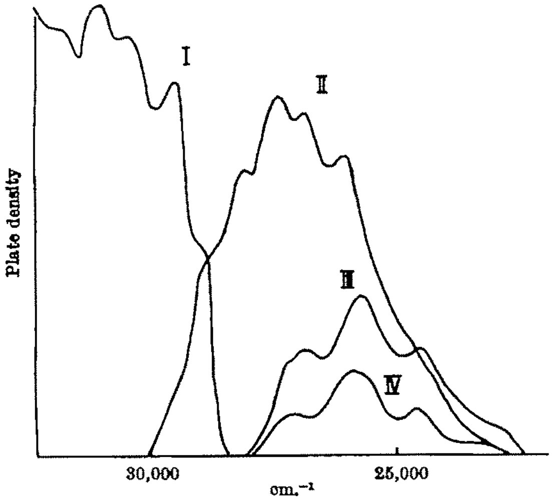

Fig. 1. Absorption (I) and emission (II) spectra of phenanthrene vapour at 9·5 mm. and 260° C. together with spectrum of delayed fluorescence (IV) and of pure anthracene vapour (III). Emission spectra were excited by the mercury 313 mμ line in each case

quartz instrument and hydrogen discharge lamp were used to photograph the absorption spectrum of the vapour at the same pressure and temperature.

Microdensitometer tracings of the plates obtained are shown in Fig. 1, together with the fluorescence spectrum of anthracene vapour excited by the 313 mμ line[5]. The delayed spectrum IV coincides in peak position and relative peak intensity with that obtained by Dikun and with the fluorescence spectrum of anthracene III; clearly it is not identical with the unsectored spectrum II and in so far as the fluorescence spectrum is a molecular characteristic it must be attributed to anthracene present as impurity.

If it is assumed that the anthracene in our phenanthrene sample is completely vapourized, its maximum possible pressure under the conditions described is $3{\cdot}8 \times 10^{-4}$ mm., corresponding to an optical density of $\sim 10^{-5}$ at 313 mμ. Thus collisional sensitization of anthracene by excited phenanthrene molecules is extremely unlikely, as is any mechanism involving the absorption of incident radiation by anthracene. We suggest, therefore, that anthracene is sensitized by collision by the excited phenanthrene dimer, which is simultaneously quenched; the lifetime of this dimer in the absence of anthracene is not known, but cannot be less than a millisecond, and in consequence the half-quenching pressure of anthracene required is at the most some 10^{-4} mm., which is within the estimated limit of impurity concentration. The absence of phenanthrene bands from the spectrum of the slow component IV suggests either that the excited dimer does not dissociate or that the quenching process is extremely efficient; this would be expected on the grounds of spin conservation since the excited dimer is in a singlet state[6].

A further requirement for efficient transfer concerns the coincidence of energy-levels in the donor and acceptor, and in this case affords an estimate of the maximum heat of formation E of the dimer from normal and excited molecules. Since the energy transferred is at least 26,400 cm.$^{-1}$ and the first excited singlet state of phenanthrene lies 28,300 cm.$^{-1}$ above the ground-state[7]:

$$E \leqslant 5{\cdot}4 \text{ kcal.}$$

which may be compared with the value ~ 6 kcal. for the heat of dissociation of the excited anthracene dimer found from the temperature dependence of self-quenching (ref. 8 and Stevens, B., and McCartin, P. J., unpublished work).

The marked resemblance between the unsectored phenanthrene spectrum presented by Dikun and the anthracene vapour fluorescence spectrum is indeed surprising. An optical density of 0·3 at 365 mμ quoted by this author suggests that the anthracene concentration in his sample is sufficient for its fluorescence spectrum to be excited directly, while the absence of phenanthrene bands could be due to complete self-quenching at the higher pressures used in his investigations.

Finally, it may be noted that if the proposed mechanism is correct, the delayed spectrum is virtually the molecular fluorescence spectrum of anthracene. This arises from a fortuitous combination of circumstances whereby a measurable fluorescence intensity of anthracene vapour at a concentration not exceeding 10^{-8} mol./l. is sensitized by an exceptionally long-lived species which is a secondary product of absorption in a region that does not overlap the emission spectrum of the acceptor.

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[1] Dikun, P. P., *Zhur. Ekspti. i Teoret. Fiz.*, **20**, 193 (1950).
[2] Williams, R., *J. Chem. Phys.*, **28**, 577 (1958).
[3] Clar, E., *Ber.*, **65**, 851 (1932).
[4] Schnepp, O., and McClure, D. S., *J. Chem. Phys.*, **20**, 1375 (1952).
[5] Stevens, B., and Hutton, E., *Mol. Phys.* (in the press).
[6] Hoijtink, G. J., paper presented at Symposium on Fast Reactions in Solution, Hahnenklee, Sept. 1959.
[7] Klevens, H. B., and Platt, J. R., *J. Chem. Phys.*, **17**, 470 (1949).
[8] Hardtl, K. H., and Scharmann, A., *Z. Naturforsch.*, **12a**, 715 (1957).

BIOPHYSICS

Sedimentation of Deoxyribonucleic Acid isolated under Low Hydrodynamic Shear

Bacteriophage deoxyribonucleic acid in solution was recently demonstrated to be degraded by moderate hydrodynamic shear gradients[1]. This observation provoked the question whether the 6–8,000,000 molecular weight, usually observed in purified preparations of deoxyribonucleic acid from animal or bacterial sources, is in any way indicative of the size of the molecules of the acid in the living cell, since in none of the common methods used for purifying deoxyribonucleic acid are precautions taken to minimize shearing.

Anderson *et al.*[2] attributed to deoxyribonucleic acid the high, shear-labile viscosity they detected in strong salt solutions of nuclear material. Unfortunately, such solutions contain protein, which could cross-link deoxyribonucleic acid. This communication briefly reports the isolation from trout sperm of shear-sensitive purified deoxyribonucleic acid of high sedimentation coefficient.

Fish sperm were used because the nuclei contain no ribonucleic acid and are easily isolated by plasmolysis and centrifugation[3]. Mr. B. Hazen of the U.S. Fish and Wildlife Service, Nashua, New Hampshire, generously provided several samples of freshly shed milt.

A number of experiments, which will be described elsewhere, suggested that although disrupting a suspension of fresh sperm nuclei by sodium dodecyl sulphate[4] or strong salt produces an extraordinarily viscous solution, relatively little of the deoxyribonucleic acid is molecularly dispersed; instead, it appears likely that the deoxyribonucleic acid occurs in the form of extremely long, tangled molecules. To obtain deoxyribonucleic acid free of occluded protein it was necessary to degrade these long molecules by applying a small shearing force.

The following preparative technique was used: A dispersion of nuclei (about 0·1 mgm. nitrogen per ml.) in 0·01 *M* sodium chloride was mixed with 1/11 volume of 5 per cent sodium dodecyl sulphate in 45 per cent ethanol[4]. After standing for 1 hr. at room temperature the solution was passed twice through a 60-cm.-long glass capillary tube, applying a calculated mean shear of 800 sec.$^{-1}$. One-third volume of 4 *M* sodium chloride was added, and after gentle shaking for a further hour, the solution was centrifuged 1 hr. at 18,000*g*. Deoxyribonucleic acid (nitrogen/phosphorus ratio 1·70) was precipitated from the water-clear supernate by ethanol. About 25 per cent of the acid in the sperm was recovered. This presumably reflects the efficiency of dispersing the deoxyribonucleic acid; by applying higher shear forces to the solution the yield of acid increased, but the mean sedimentation coefficient of the product fell.

The secondary structure appeared to be intact since the deoxyribonucleic acid showed a 30 per cent hyperchromicity at 260 mμ on alkalination[5]. For physical study the deoxyribonucleic acid was redissolved in 0·01 *M* sodium chloride under gentle agitation.

The ultracentrifuge techniques have been previously described[1]. The cells were filled at a mean shear-rate of 1,000 sec.$^{-1}$. A typical distribution of the sedimentation coefficients in one of these preparations is shown in Fig. 1. The sedimentation diagram was unchanged after treatment with versene, trypsin, or chymotrypsin. Fig. 1*C* shows the corresponding distribution in deoxyribonucleic acid isolated from the same batch of milt by the method of Kay, Simmons, and Dounce[4].

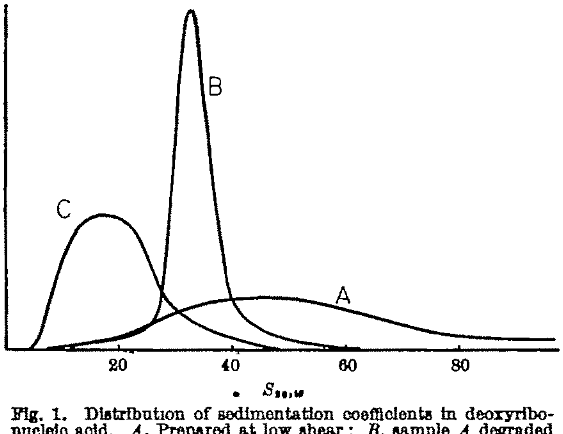

Fig. 1. Distribution of sedimentation coefficients in deoxyribonucleic acid. *A*, Prepared at low shear; *B*, sample *A* degraded by shear; *C*, prepared by method of Kay *et al.* (ref. 4). Solvent: 0·2 *M* sodium chloride, 0·015 *M* sodium citrate

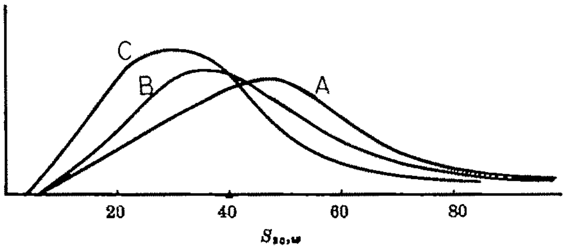

Fig. 2. Distribution of sedimentation coefficients in deoxyribonucleic acid. *A*, Intact; *B*, sheared once; *C*, sheared thrice. Solutions heated before centrifugation. Solvent: 0·2 *M* sodium chloride, 0·015 *M* sodium citrate. Ordinate scale twice that in Fig. 1

Deoxyribonucleic acid prepared in the manner outlined above and sheared at 40,000 sec.$^{-1}$ is shown in Fig. 1*B*. The shearing lowered the viscosity of the solution 25 per cent (as measured in a simple Ubbelohde viscometer at relatively high shear). When the solution was sheared twice more, the peak appeared even sharper, and the maximum dropped about 2 *S* (Svedberg units), while the viscosity dropped a further 10 per cent. Two anomalies are apparent in these results: (1) the viscosity changes are disproportionate to the drastic change in the sedimentation diagram on the first shearing, and the small change afterwards; (2) if the mechanism of the shear degradation is that previously discussed[1] the shearing should either halve the molecule or leave it intact, and it is unlikely that a drop from 32 *S* to 30 *S* represents a halving of the molecular weight.

However, the width of the sheared diagrams increased with dilution, and it appeared probable that the boundary was self-sharpened. This effect was not obviated at extreme dilution, but disappeared when the solutions were heated.

Briefly heating deoxyribonucleic acid solutions of moderate ionic strength to 100° C. destroys the ordered helical configuration of the molecule[6] and broadens the sedimentation distribution diagram[7], usually without decreasing the molecular weight. The broadening of the sedimentation diagram probably reflects an increased polydispersity of shape, together with some molecular degradation and some aggregation through re-formed hydrogen bonds[8]. Since it is unlikely that aggregation and degradation could fortuitously counterbalance if either process occurred to a great extent, it seems probable that at least the major features of the distribution of the sedimentation coefficients are preserved in the heated preparations.

Fig. 2 depicts the sedimentation diagrams of sheared deoxyribonucleic acid samples after heating. The fact that these distributions appear qualitatively consistent with the viscosity studies supports the suggestion that the diagrams approximate the molecular size distribution which was previously concealed in the hypersharp boundaries.

Under the brief shearing a fraction of the susceptible molecules escaped degradation each time. The only unexpected feature of the diagrams was a persistence of the 'tail' on the high *S* side. Since this material was not shear-labile, it might comprise aggregates of low asymmetry, possibly artefacts produced by the heating.

A number of observations indicated that self-sharpening occurs whenever the change of concentration (dc/dr) at a point within the boundary is sufficiently high. Thus, on shearing, the scission of the larger molecules increased dc/dr on the low *S* side of the boundary, giving rise to an artificial sharpness which over-emphasized the effect of the shearing.

In summary, it has been shown that deoxyribonucleic acid of unusually high sedimentation coefficient can be prepared. If these results can be extended to deoxyribonucleic acid from somatic tissues, it appears to me that at present no limit can be set to the length of the molecule of deoxyribonucleic acid *in vivo*. Protein bridges may exist along these 'molecules', but, until some evidence for them is forthcoming, it is simpler to imagine that the double helix persists uninterruptedly—perhaps for the length of a chromosome[9].

I am indebted to Prof. Francis O. Schmitt and Prof. Cyrus Levinthal for valuable discussions in the course of this work, and to Mrs. K. Y. Ho for technical assistance.

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[1] Davison, P. F., *Proc. U.S. Nat. Acad. Sci.*, **45**, 1510 (1959).

[2] Anderson, N. G., *Nature*, **172**, 807 (1953). Fisher, W. D., Anderson, N. G., and Wilbur, K. M., *Exp. Cell. Res.*, **18**, 100 (1959).

[3] Felix, K., Fischer, M., and Krekels, A., "Prog. in Biophys.", **6**, 1 (1956).

[4] Kay, E. R. M., Simmons, N. S., and Dounce, A. L., *J. Amer. Chem. Soc.*, **74**, 1724 (1952).

[5] Hotchkiss, R. D., in "Methods in Enzymology", **3**, 708, edit. by Colowick, S. P., and Kaplan, N. O. (Academic Press, New York, 1957).

[6] Rice, S. A., and Doty, P., *J. Amer. Chem. Soc.*, **79**, 3937 (1957). Geiduschek, E. P., and Holtzer, A., "Adv. Biol. Med. Phys.", **6**, 431 (1958).

[7] Shooter, K. V., Pain, R. H., and Butler, J. A. V., *Biochim. Biophys. Acta*, **20**, 497 (1956). Schachman, H. K., *J. Cell. Comp. Physiol.*, Supp. 1, 71 (1957).

[8] Doty, P., Boedtker, M., Fresco, J. R., Haselkorn, R., and Litt, M., *Proc. U.S. Nat. Acad. Sci.*, **45**, 482 (1959).

[9] Levinthal, C., Josiah Macy, Jr., First Conference on Genetics, 1959 (in the press).

Lubrication of Mammalian Joints

THE experiments and arguments of McCutchen[1] and Lewis and McCutchen[2] appear to confirm the theory of lubrication of mammalian joints first put forward twenty-seven years ago with particular reference to the intra-articular fibrocartilages and later developed in connexion with particular problems of articular structure, general and special[3]. The essence of that theory is that there is a sensibly thick film of synovial fluid between articular surfaces while the joint is in motion, and that this fluid is not only maintained between the surfaces but also flows between them in the direction of motion in accordance with the laws of viscous hydrodynamics. The question of the origin of the fluid was left in abeyance, being a physiological rather than a mechanical problem.

It is an axiom, too often unstated, that the flow of lubricant film while it is acting requires that the fluid shall adhere to the fixed and the moving surfaces; this is tantamount to saying that these surfaces shall be thoroughly soaked with fluid. Movement of one of the surfaces will then necessarily produce a laminar flow if the fluid be viscous, as is easily demonstrated by simple physical models at speeds commensurate with those found in our joints. The structural basis for the sponge-like character of articular cartilage has already been shown by myself[4]. This structure differs markedly from that described by Benninghoff, referred to in the communication from Lewis and McCutchen. The late Prof. Benninghoff told me verbally that he had not examined articular cartilage by the methods given in the preceding reference. Lewis and McCutchen have shown that the articular sponge not only soaks up fluid, thus ensuring its adhesion to the bearing surfaces, but also permits it to be squeezed out, thus ensuring an instantaneous formation of a lubricant film which can then itself be moved as stated above. Such a rapid formation of lubricant fluid is essential in the joints where the range of movement is small by engineering standards and also alternates rapidly in direction. It should be stressed, however, that any theory of articular lubrication must cover all the phenomena of structure within the joint cavity, and should account for the presence, shape and disposition of soft structures (for example, fatty pads) and ancillary deformable structures (for example, articular disks) found in certain joints where the general theory of viscous lubrication indicates a need for their presence. The 'weeping lubrication' spoken of by Lewis and McCutchen appears to complete the existing theory rather than to supplant it, and is a valuable addition to our knowledge of biomechanics.

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[1] McCutchen, C. W., *Nature*, **184**, 1284 (1959).

[2] Lewis, P. R., and McCutchen, C. W., *Nature*, **184**, 1285 (1959).

[3] MacConaill, M. A., *J. Anat.*, **66**, 210 (1932); *Proc. Roy. Irish Acad.*, **50**, B, 7 (1944); *J. Bone and Jt. Surg.*, **32**, B, 244 (1950). Davies, D. V., *J. Anat.*, **82**, 9 (1948). Bruce, J., and Walmsley, R., *Brit. J. Surg.*, **25**, 17 (1937); *J. Anat.*, **72**, 260 (1938). Jones, E. S., *Lancet*, i, 426 (1934); *Lancet*, i, 1043 (1936).

[4] MacConaill, M. A., *J. Bone and Jt. Surg.*, **33**, B, 251 (1951).

WHILE we agree with Prof. MacConaill that there is a thick layer of fluid over most of the area between the articulating surfaces of a joint, we none the less insist that how it gets there is a mechanical rather than a physiological problem. When a joint is loaded, the fluid between the surfaces will be at high pressure, and this must be overcome by whatever process supplies the fluid. Weeping lubrication solves the problem by automatically pressurizing the reservoir from which the fluid is supplied. This is accomplished at the expense of gentle contact at a few high spots in order to press the sponge-like cartilages back into the fluid they are soaked with.

Hydrodynamic lubrication can, in principle, drag fluid into the high-pressure region, but the amount is very small. A simple calculation[1,2] for the knee joint of a sheep shows that, even were the shapes and motion of the joint surfaces ideal, the amount of fluid which can be brought between the articulating surfaces in one swing of the leg at walking speed is equivalent to a layer no greater than half a micron thick. We have shown that the weeping process can provide 30–70 times this amount of fluid; so it seems unlikely that the hydrodynamic contribution is very important. At lower speeds, hydrodynamic lubrication seems even less likely, because the amount of fluid supplied per swing follows the half-power of the sliding speed. If one follows the hydrodynamic theory down to extremely low sliding velocities in the hope of being helped by the unusual properties of synovial fluid[3], one finds that there is no appreciable increase in viscosity of the lubricant until the sheep walks at less than 10^{-7} m.p.h., at which speed the lubricating film is less than 10 Å. thick. Few surfaces are this smooth, and in practice, boundary lubrication conditions would set in at far higher speeds.

Failure of the lubricant film at low sliding speeds, which is a standard feature of hydrodynamic lubrication[3], ought to lead to a high coefficient of starting friction. Charnley[4] actually measured starting friction and found that no more force was required to start a joint sliding than to keep it sliding.

The precise structure of the cartilage is not of prime importance. What is important is that it should act as a sponge in which fluid can move easily in a direction normal to the articular surface; that this does occur is confirmed by the experiments detailed in the second half of our original communication. It is also important that the lateral permeability should not be too high, otherwise the cartilage would rapidly collapse when the load was applied. Our experiments gave no indication of a high lateral permeability, but this aspect of our hypothesis does require more detailed, preferably quantitative, study.

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[1] Kearton, W. J., "Steam Turbine Theory and Practice" (Pitman, London, 1951).

[2] Ogston, A. G., and Stanier, J. E., *J. Physiol.*, **119**, 244 (1953).

[3] Bowden, F. P., and Tabor, D., "The Friction and Lubrication of Solids" (Oxf. Univ. Press, 1954)

[4] Charnley, J., Inst. Mech. Eng., Symp. on Biomechanics, April 17, 1959.

BIOCHEMISTRY

Preparation and Some Properties of a Soluble Ribonucleic Acid in Yeast

The function of soluble ribonucleic acid in the transfer of activated amino-acids to the microsomal ribonucleoprotein particles has been correlated to the presence of a specific terminal *–pCpCpA* group[1–3], and the occurrence of pseudouridylic acid in the molecules[4]. One of the basic requirements for the full understanding of the structural basis of the function of soluble ribonucleic acid would be the preparation of a ribonucleic acid sample which is homogeneous, unaltered, and having a functional activity not different from that present in the living cells. The principal purpose of this study has been to establish the technique whereby a sample of acid which satisfies the above requirements is prepared. The attempt towards this direction led to preparation of an active ribonucleic acid sample which is homogeneous when judged by electrophoretic examination and sedimentation analyses

It has been reported that in yeast cells the bulk of the active ribonucleic acid is localized in the fraction soluble at *p*H 5·0 or *p*H 5·3 of the 105,000*g* supernatant of the homogenate[4]. A crude sample of the acid obtained by the phenol method of Kirby[5] from the *p*H 5·0 supernatant fraction was contaminated with polysaccharide which has, however, little effect on the incorporation of leucine labelled with carbon-14 into the ribonucleic acid under the conditions previously described[4]. The sample of crude acid was dissolved in 0·001 *M* potassium phosphate buffer at *p*H 7·1, and shaken with 'Ecteola'-cellulose[6] for 5 min. in the cold. About 90 per cent of ultra-violet-absorbing substance originally present in the crude ribonucleic acid preparation was adsorbed on 'Ecteola', whereas all polysaccharides remained uncombined. The 'Ecteola'–ribonucleic acid complex was thoroughly washed with the buffer to remove the unadsorbed materials. The bulk of the ultra-violet-absorbing substance could then be released with 0·3 *M* sodium perchlorate at *p*H 7·1, and recovered by ethanol precipitation. Under these conditions, the ribonucleic acid prepared from the cytoplasmic ribonucleoprotein particles was held firmly to 'Ecteola' and did not come out with 0·3 *M* sodium perchlorate. About 75 per cent of ultra-violet-absorbing substance in the original crude sample of ribonucleic acid was recovered by precipitation by ethanol. Electrophoretic examination revealed the removal of polysaccharides. However, a small peak was detectable slightly ahead of the main acid peak. This fast component was removed completely by 24-hr. dialysis. An electrophoretically homogeneous sample of a soluble ribonucleic acid could thus be obtained. A rapid 'Ecteola' treatment seems to be essential for the successful preparation. An extensive degradation and inactivation of the ribonucleic acid were observed, when the column chromatographic fractionation on 'Ecteola' of the ribonucleic acid was tried.

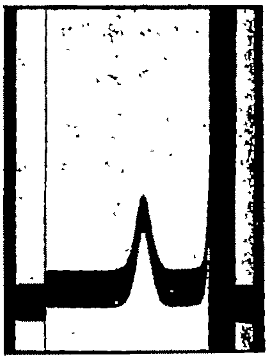

Fig. 1. Sedimentation pattern of purified yeast soluble ribonucleic acid. Picture taken after 24 min. at 56,900 rev./min. in a synthetic boundary cell. 1 6 per cent solution in 0·1 *M* sodium chloride–0·03 *M tris*(hydroxymethyl)aminomethane at *p*H 7 1

The purified sample was also homogeneous in the sedimentation analyses (Fig. 1), giving a sedimentation constant of $S^{0}_{20} = 4\cdot0$, extrapolated from the values obtained at three sample concentrations in 0·1 *M*

Table 1. Some Properties of Soluble Ribonucleic Acid compared with those of Ribonucleic Acid prepared from Cytoplasmic Ribonucleoprotein Particles of Yeast Cells (*Saccharomyces cerevisiae*)

	λ_{max}	$\varepsilon(P)$	$\lambda_{min.}$	$\varepsilon(P)$	$[\eta]_{20}$ 100 ml./gm.	S^{0}_{20}	Mol wt	Axial length a(Å.)	b(Å.)
Soluble ribonucleic acid	258	6,980	230	3,330	0 05	4 0	27,000	133	10
Ribonucleic acid from cytoplasmic ribonucleoproteins	258	7,620	231	3,100	0 54	22 (main)* 19 4 ∼ 5	1,100,000	—	—

All measurements were done in 0·1 *M* sodium chloride—0 03 *M tris*(hydroxymethyl)aminomethane at *p*H 7 1. Molecular size and shape for soluble ribonucleic acid were calculated assuming partial specific volume = 0·58, and the molecular model of a rigid ellipsoidal rod.

* Three peaks were detected. No correction for dependence on temperature and concentration.

sodium chloride – 0·03 *M tris*(hydroxymethyl)-aminomethane at *p*H 7·1. The molecular weight of the ribonucleic acid calculated from the sedimentation constant and the intrinsic viscosity ($[\eta]_{20}$ = 0·05) was 27,000 (Table 1). The nitrogen/phosphorus ratio of the product was 1·63. Contamination of deoxyribonucleic acid in the most purified sample was around 0·25 per cent relative to ribonucleic acid.

The nucleotide composition analyses by 'Dowex-1' column chromatography[7] confirmed the previous findings of the predominant localization in the soluble ribonucleic acid of pseudouridylic acid[4,8] and other minor nucleotide components[8].

The purified ribonucleic acid combined L-leucine in the proportion of 2·86 mμM/mgm. ribonucleic acid under the conditions described previously[4]. The value was about 40 per cent higher than that obtained when the crude sample of ribonucleic acid was tested. This seems to be not due to the inhibitory action of contaminated materials in the crude acid, but rather to the removal of inactive ultra-violet-absorbing substances during the purification.

Although the ribonucleic acid obtained is fairly homogeneous from the criteria here adopted, it is not known at the present time whether the acid is a mixture of ribonucleic acids each of which is specific for the attachment of individual amino-acids[9].

A full account of this work will be published later.

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[1] Hecht, L. L., Stephenson, M. L. S., and Zamecnik, P. C., *Proc. U.S. Nat. Acad. Sci.*, 45, 505 (1959).

[2] Zachau, H. G., Acs, A., and Lipmann, F., *Proc. U.S. Nat. Acad. Sci.*, 44, 885 (1958).

[3] Preiss, J., Berg, P., Ofengand, E. J., Bergmann, F. H., and Dieckmann, M., *Proc. U.S. Nat. Acad. Sci.*, 45, 319 (1959).

[4] Osawa, S., and Otaka, E., *Biochim. Biophys. Acta*, 36, 549 (1959).

[5] Kirby, K. S., *Biochem. J.*, 64, 405 (1956).

[6] Peterson, E. A., and Sober, H. A., *J. Amer. Chem. Soc.*, 78, 751 (1956).

[7] Osawa, S., Takata, K., and Hotta, Y., *Biochim. Biophys. Acta*, 28, 271 (1958).

[8] Dunn, D. B., *Biochim. Biophys. Acta*, 34, 286 (1959).

[9] Smith, K. C., Cordes, E., and Schweet, R. S., *Biochim. Biophys. Acta*, 33, 286 (1959).

Biosynthesis of Chlorophylls a and *b* in *Chlorella vulgaris*

WE have shown that both carbon atoms of glycine were incorporated into chlorophyll *a* when *Chlorella vulgaris* was incubated with ^{14}C-glycine for a period of 1 hr. During this period no carbon-14 from the carboxyl group could be detected in chlorophyll *b*, whereas the incorporation of the carbon from the methylene group was several times higher than in the case of chlorophyll *a*. Only after longer periods of incubation with 1-^{14}C-glycine was the isotope found in chlorophyll *b* as well.

The purity of the glycine samples (1-^{14}C-glycine and 2-^{14}C-glycine, specific activities about 50 μc. per mgm.) was determined chromatographically; the radioautograms contained no impurities. *Chlorella vulgaris*, strain '211/11*m*' or 'Cornell 11' (received from the Culture Collection of Algae and Protozoa, University of Cambridge), was grown in sterile liquid cultures in volumes of 100 ml. per flask. The composition and the preparation of the medium were described by Della Rosa *et al.*[1]. In order to obtain a homogeneous algal culture of the 'D_A type'[2], the Tamiya procedure of 'synchronous culture' has been adopted (16 hr. light of 1 k.lux and 8 hr. of darkness). After maximal verdancy of the cells was achieved, 10 μc. ^{14}C-glycine were introduced. Samples were then withdrawn after various periods of incubation under 1 k.lux of light. The yield per flask amounted to about 100–150 mgm. of dry algae. 'Raw chlorophyll', free of xanthophylls, was extracted from the frozen material according to Strain and Manning[3] and chromatographed on Whatman No. 1 paper impregnated with saccharose (Sporer *et al.*[4]) using Bauer's solvent[5]. Chlorophylls *a* (R_F about 0·65) and *b* (R_F about 0·35) were eluted with ethyl ether and determined spectrophotometrically, employing the absorption coefficients given by Smith and Benitez[6]. The wet combustion of the chlorophyll samples was carried out as described by Van Slyke and Folch[7], using inactive glucose as carrier, and the carbon dioxide converted into barium carbonate. The labelled carbon dioxide set free from the barium carbonate was measured in the gas Geiger counter of Broda *et al.*[8] and calculated to 100 per cent counting yield. The results are presented in Table 1.

Table 1. INCORPORATION OF CARBON-14 FROM GLYCINE INTO CHLOROPHYLLS *a* AND *b*

Substrate	Time of incubation (hr.)	Specific activity of chlorophyll (c.p.m. × 10^{-4} per mgm. C)					
		a			*b*		
		No.	Results	Average	No.	Results	Average
1-^{14}C-glycine	1	I	1·29	1·02	I	0	0
		II	0·91		II	0	
		III	0·85		III	0	
	6	I	4·96	5·25	I	5·36	6·16
		II	5·17		II	6·71	
		III	5·62		III	6·43	
2-^{14}C-glycine	1	I	0·59	0·68	I	3·09	2·41
		II	0·75		II	—	
		III	0·70		III	1·73	
	6	I	8·80	9·88	I	21·8	15·8
		II	6·55		II	9·72	
		III	12·8		III	—	

Della Rosa *et al.*[1] found in experiments in which the period of incubation lasted several days that, in contrast to the biosynthesis of hæm, not only the methylene carbon but also the carboxyl carbon of glycine is utilized in the synthesis of chlorophylls *a* and *b*. In view of the fact that the isotope concentrations in both chlorophylls were of the same order of magnitude they decided against the current theory that chlorophyll *b* may be derived from chlorophyll *a*. Since our results after 6 hr. of incubation are similar to those of Della Rosa *et al.*, and since after 1 hr. of incubation (that is to say, in the beginning stages of biosynthesis) the uptake of carbon-14 from the two sources is different for the two chlorophylls, the suggestion that the path of biosynthesis of chlorophylls *a* and *b* may be different seems quite reasonable.

It would appear from our work that in the biosynthesis of chlorophyll *b*, as in the synthesis of hæm, the methylene carbon of glycine plays the main part, and that this process may be simpler or quicker than the incorporation of this carbon into chlorophyll *a*. On the other hand, both carbon atoms are utilized equally well by chlorophyll *a*. The scheme proposed by Della Rosa *et al.*, namely, that the carboxyl carbon is incorporated into the chlorophylls via the citric acid cycle, cannot be accepted, as in this case the carbon-14 concentration in chlorophylls *a* and *b* after 1 hr. of incubation should be of similar magnitude. As can be seen from Table 1, there is considerable incorporation of the carboxyl carbon into chlorophyll *a* and no incorporation at all into chlorophyll *b* after 1 hr. of incubation.

After 6 hr. the specific activities of both chlorophylls are of the same order, and are always higher on incubation with 2-^{14}C-glycine than with 1-^{14}C-glycine. This observation may suggest some incorporation of the carboxyl carbon of glycine, which takes no direct part in the biosynthesis of chlorophyll *b*, into some other group which may then be equally well used in the synthesis of both chlorophylls.

We are indebted to Prof. E. Broda in Vienna, where our work was started, and to Prof. I. Reifer in Warsaw, where our work was concluded, for helpful discussions.

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[1] Della Rosa, R. J., Altman, K. I., and Salomon, K., *J. Biol. Chem.*, **202**, 771 (1953).

[2] Tamiya, H., "Ann. Rev. Plant Physiol.", **8**, 309 (1957).

[3] Strain, A. H., and Manning, W. M., *J. Biol. Chem.*, **144**, 625 (1942).

[4] Sporer, A. H., Freed, S., and Sancier, K. M., *Science*, **119**, 68 (1954).

[5] Bauer, L., *Naturwiss.*, **39**, 88 (1952).

[6] Paech, K., and Tracey, M. V., "Modern Methods of Plant Analysis", **4**, 142 (1955).

[7] Van Slyke, D. D., and Folch, J., *J. Biol. Chem.*, **136**, 509 (1940).

[8] Broda, E., and Rohringer, G., *Z. Elektrochem.*, **58**, 634 (1954).

Stereospecificity of Enzymic Reactions involving Mevalonic Acid

MOST of the biosynthetic studies with mevalonic acid have so far been carried out with the racemic substance labelled with carbon-14. There have been no reports indicating a utilization greater than 50 per cent of DL-mevalonate either *in vivo* or *in vitro* in any enzymic reaction. Thus, Tavormina, Gibbs and Huff[1], who made the discovery that mevalonate was an outstanding precursor of cholesterol, noted that about 43 per cent of the DL-[2-^{14}C] mevalonate was converted in liver homogenates into cholesterol; an observation confirmed by Popják *et al.*[2]. Gould and Popják[3] found that of the dose of carbon-14 contained in DL-[2-^{14}C] mevalonate and injected into mice about 40 per cent appeared in body sterol, 10 per cent in expired carbon dioxide and 50 per cent in the urine. Both Tchen[4] and Levy and Popjak[5,6] observed that in the mevalonic kinase reaction, in the phosphorylation of mevalonate with adenosine triphosphate to 5-phosphomevalonate, no more than 50 per cent of the DL-mevalonate was used by the enzyme. All these observations led to the conclusion that only one enantiomorph of mevalonate could act as substrate in biosynthetic reactions, and this was assumed to be the natural (+) isomer, $[\alpha]^{24}_{D}$, + 10° (Wolf *et al.*[7]).

We have obtained direct evidence on this question and found that the mevalonate excreted in the urine of animals given DL-mevalonate is of the unnatural optical configuration, and that the 5-phosphomevalonate, prepared enzymically with liver mevalonic kinase[5,6] was an optically active substance.

Three rats, kept in metabolism cages, were given daily in their food 50–100 μmoles of potassium DL-mevalonate mixed with a small amount of DL-[2-^{14}C] mevalonate. The total dose given to the three animals was 201 mgm. (calculated as mevalonic acid lactone). The urine, which was collected and pooled, was brought to *p*H 9 and extracted with chloroform for 20 hr. to remove neutral material; then it was made acid to congo red and extracted again for 20 hr. with chloroform. Nearly all the carbon-14 contained in the urine was recovered in this extract. The second chloroform extract was evaporated under vacuum and the residue distributed between water and ether. The aqueous layer was evaporated to dryness under reduced pressure at 30–40° and the residue distilled at 0·001 mm. of mercury pressure, the bath temperature being raised to 130°. The distillate (105 mgm.) was refluxed with diphenylmethylamine in benzene for 1½ hr. and mevalonic acid diphenylamide (149 mgm.) isolated as described previously[8]. On recrystallization from benzene, 105 mgm. of the amide (m.p. 95–96°) were obtained with $[\alpha]^{24}_{589}$ + 2·2° in ethanol. The results of optical rotatory dispersion measurements, made at a concentration of 1·2 gm./100 ml. in ethanol, are shown in Fig. 1.

Wolf *et al.*[7] gave $[\alpha]^{24}_{D}$ − 2·0° (in ethanol) for the diphenylamide of mevalonic acid lactone isolated from distillers' solubles. It is apparent from the results that the rats have utilized the natural and excreted the unnatural isomer of mevalonic acid.

5-Phosphomevalonate was prepared, as the barium salt, from DL-mevalonate with liver mevalonic kinase according to the procedure of Levy and Popják[6]. Optical rotatory dispersion measurements were made on an aqueous solution (0·25 gm./100 ml.) of 5-phosphomevalonic acid (after removal of barium); these results are shown in Fig. 2. The specific rotation at 589 mμ was $[\alpha]^{24}_{589}$ − 6·1° and the molecular rotation $[M]^{24}_{589}$ − 13·9°. These observations leave no doubt that the former assumptions as to the specificity of

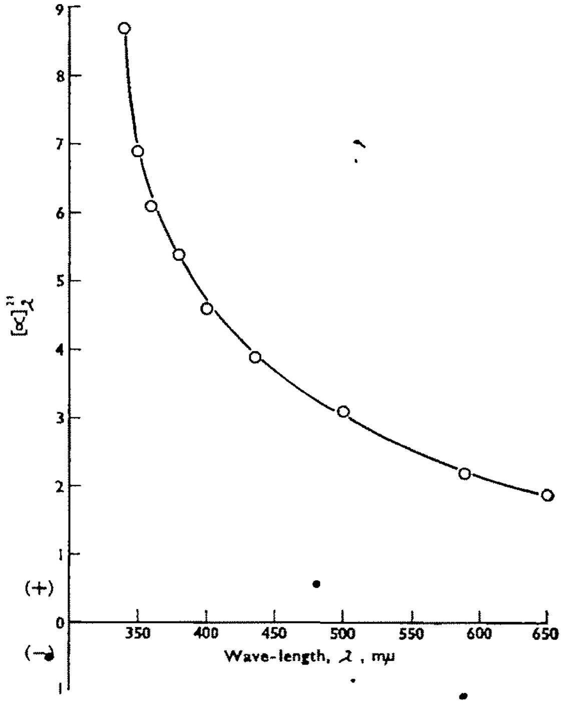

Fig. 1. Optical rotatory dispersion data on the diphenylmethylamide of mevalonic acid in ethanol. The ordinates give specific rotations $[\alpha]^{24}_{\lambda}$

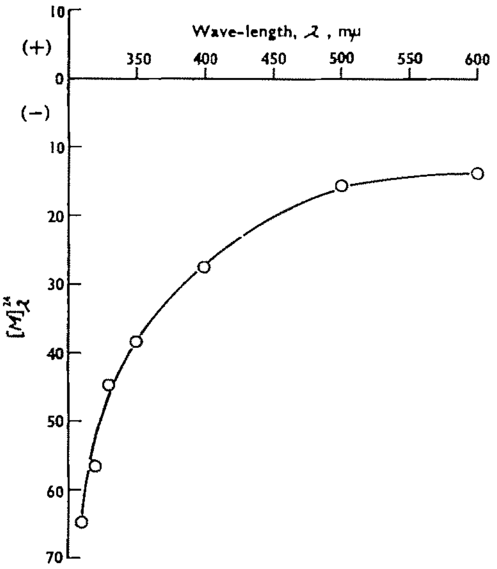

Fig. 2. Optical rotatory dispersion results on 5-phosphomevalonic acid in water. The ordinates give the molecular rotations $[M]^{24}_{\lambda} = [\alpha]^{24}_{\lambda} \times$ mol. wt./100

enzymic reactions for one optical isomer of mevalonic acid are correct.

We are indebted to Dr. C. Sweeley of the National Heart Institute, National Institutes of Health, Bethesda, Md., and to Dr. W. Klyne of the Postgraduate Medical School of London, for the optical rotatory dispersion measurements on the diphenylamide and on the 5-phosphomevalonic acid respectively.

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[1] Tavormina, P. A., Gibbs, M. H., and Huff, J. W., *J. Amer. Chem. Soc.*, 78, 4498 (1956).
[2] Popják, G., Gosselin, L., Gore, I. Y., and Gould, R. G., *Biochem. J.*, 69, 238 (1958).
[3] Popják, G., and Gould, R. G., *Biochem. J.*, 66, 51*P* (1957).
[4] Tchen, T. T., *J. Biol. Chem.*, 233, 1100 (1958).
[5] Levy, H. R., and Popják, G., *Biochem. J.*, 72, 35*P* (1959).
[6] Levy, H. R., and Popják, G., *Biochem. J.* (in the press).
[7] Wolf, D. E., Hoffman, C. H., Aldrich, P. E., Skeggs, H. R., Wright, L. D., and Folkers, K., *J. Amer. Chem. Soc.*, 78, 4499 (1956).
[8] Cornforth, J. W., Cornforth, R. H., Popják, G., and Gore, I. Y., *Biochem. J.*, 69, 146 (1958).

Glucose-isomerase in Barley Malt

WORK in this Institute on the enzymatic breakdown of starch has included above all the qualitative and quantitative determination of products resulting from this breakdown[1]. We have observed that in every case fructose occurred beside the glucose derivatives already in the barley malt extract[2]. The presence of glucose may be attributed to the activity of the amylo-glucosidase enzyme of the barley malt. Among the glucose derivatives maltose is produced by the activity of the well-known β-amylase. The oligosaccharides, from which we succeeded in identifying altogether eight members in two homologous series, beginning with maltotriose and finishing with malto-octaose, were produced by the activity of the amylase. Besides the amylolytic enzymes, there must also be a glucose-isomerase enzyme in action, and it must be its effect which results in the appearance of fructose.

We have verified the presence of fructose by paper chromatography. Fructose is well characterized by the lemon-coloured spot formed in *p*-amino-phenol[3] used as detecting agent. This spot appears only when fructose is present. Among the other carbohydrate products yielded by the breakdown of starch, maltose gives in *p*-amino-phenol an ochre-yellow spot while the oligosaccharides produce spots in colours ranging from reddish yellow to reddish brown. Very characteristic are further for fructose the R_F and R_G values respectively, which could be exactly determined by the control test. In this way the presence of fructose was undoubtedly proved. The quantity of fructose introduced by the malt-extract, and the fructose and saccharose content of the raw material were exactly determined, and it could be observed that at the end of the saccharification process the reaction-mixture contained an amount of fructose which was two or three times higher than that of the raw material. This could be observed no matter whether pure starch of some starch-containing raw material (potatoes or corn) had been chosen for the saccharification. This cannot occur except in the case when, during the breakdown of the starch, enzyme action transforms glucose into fructose. The presence of such an enzyme was proved experimentally; thus we treated a 5 per cent glucose solution with barley malt extract and found that during the reaction the amount of glucose decreased while that of the fructose increased.

We treated a 13–17 days-old malt germinated on a threshing floor with a 5 per cent solution of analytical glucose at 55° C. and 5·2 *p*H, during 2 hr. The carbohydrates were quantitatively determined in both solutions before mixing and in the reaction products after the treatment, using paper chromatography.

The results obtained by quantitative analysis of the chromatogram spots are given in Table 1, every result being for 150 ml. of the solutions.

Table 1

Colour of the spot	$R_F \times 100$	Identified quality of the spot	1*	2† (gm.)	3‡
Lemon-yellow	26 8	Fructose	—	0·76	0·23
Dark brown	24·0	Glucose	4·6	3·92	0 26
Light brown	18·6	Saccharose	—	0·06	0 06
Ochre-brown	15 2	Maltose	—	0 78	0·68
Reddish-brown	11·7	Maltotriose	0 2	0 40	0·36
Yellowish-brown	—	Oligosaccharides	—	0 52	0·58
Brown	—	Dextrins	—	0 92	0·81
		Totals	4·8	7·45	2·98

* Mixture of 100 ml. 5 per cent glucose solution and 50 ml. distilled water.

† 100 ml. 5 per cent glucose solution and 50 ml. barley malt extract.

‡ Mixture of barley malt extract and 100 ml. distilled water has been chromatographed and analysed.

These results show that, due to the barley malt extract, a quantity of about 1 gm. disappeared from the glucose in solution while the amount of the fructose increased by about 0·5 gm.

Thanks are due to Mrs. Agnes Osztróvszky and Miss Eva Gábor for their kind assistance.

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[1] Bártfay, J., and Vámos, Mrs. L., *Die Branntweinwirtschaft*, 6, 188 (1959).

[2] Bártfay, J., *Die Branntweinwirtschaft*, 21, 517 (1959).

[3] Vámos, Mrs. L., *Magyar Kémiai Folyóirat*, 8, 253 (1958).

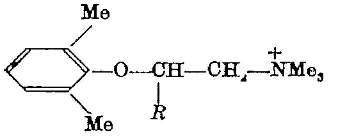
ANIMAL PHYSIOLOGY

Powerful Adrenergic Neurone-blocking Agents related to Choline 2,6-Xylyl Ether Bromide

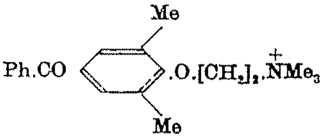
THE discovery that the functioning of the post-ganglionic sympathetic adrenergic nerves could be impaired by choline 2,6-xylyl ether bromide (I), *TM* 10 (refs. 1, 2), led to a search for compounds having a similar effect, but lacking the parasympathomimetic actions which preclude its use in man[3]. Such a selective blocking action was found in a series of benzyl quaternary ammonium salts[4], and one of these, bretylium tosylate, was eventually shown to be a valuable antihypertensive agent in man[5]. This compound can depress conduction in various nerves, and the specificity of its blocking effect on the sympathetic nervous system seems to be related to its selective accumulation in adrenergic neurones (unpublished work).

Investigation of analogues of choline 2,6-xylyl ether bromide showed that the β-methyl substituted compound (II) had a similar but more specific action on the adrenergic nerve mechanism[6]. We also have prepared many compounds related to *TM* 10 and screened them for adrenergic neurone-blocking action in the cat, by their ability to relax the nictitating membrane without causing mydriasis. We have found that additional substitution of *TM* 10 in the 4-position by ethoxycarbonyl, acetyl or benzoyl, leads to reduction or abolition of the gross parasympathomimetic effects, with retention or enhancement of the antiadrenergic properties.

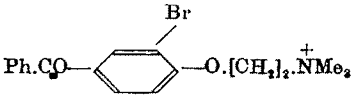
One of the most active of these compounds, 172*C*58, is 10–20 times more active than *TM* 10 and is virtually devoid of parasympathomimetic properties. The corresponding bromo-analogue, 225*C*59, has similar adrenergic neurone-blocking properties, but produces muscarine-like effects at high dose-levels. Many chemical variations on the compound 172*C*58, which included branching or lengthening the chain, additional substitution or hydrogenation of the 4-benzoyl group and several variations of the groups in the 2- and 6-positions and in the cationic head, have led to lower activities. Related compounds are being investigated.



I, *R*=H, II, *R*=Me



172*C*58*



225*C*59*

* Patents pending

We examined 172*C*58, as its bromide, by the same methods as were used with bretylium[7]. In cats, intravenous doses of 0·05–0·5 mgm./kgm. rapidly depressed for several hours the responses to stimulation of various adrenergic nerves; for example, the increase in heart-rate caused by stimulation of the inferior cardiac nerve, the relaxation of the uterus caused by stimulation of the hypogastric nerve and the contraction of the nictitating membrane during stimulation of the postganglionic cervical sympathetic trunk. These doses lowered the blood pressure of cats in the supine position and caused marked postural hypotension. These effects occurred without depression of the responses to injected noradrenaline or adrenaline; indeed their actions were enhanced, as occurs after sympathectomy. Sweating caused by sympathetic nerve stimulation in the cat was not abolished. Since the postganglionic fibres to the sweat glands are cholinergic, this suggests that the action of the compound is largely confined to adrenergic neurones. The compound showed weak and transient ganglion-blocking properties. No blocking action on the mechanism for release of pressor amines from the adrenals was detected in acute experiments. Neuromuscular paralysis of voluntary muscle occurred with doses about 50 times greater than those which affected the adrenergic nerve mechanism.

The distribution in the tissues of the drug labelled with carbon-14 on one of the quaternary methyl groups showed that it had a high affinity for the adrenergic neurones. 2 hr. after the injection of 1 mgm./kgm. subcutaneously in the cat, the radioactivity in sympathetic ganglia and adrenergic nerve trunks indicated the presence of concentrations of approximately 0·005 and 0·002 μmole/gm., respectively—some five times the highest found in other ganglia and nerves. Furthermore, when the drug was applied topically to adrenergic nerves between stimulating electrodes and a suitable end-organ, the response of the latter was abolished. This was shown in tests using the hypogastric nerve, and the sympathetic fibres of the greater auricular and mesenteric nerves in the rabbit, and also on the postganglionic cervical sympathetic and inferior cardiac nerves of the cat. Like bretylium, the drug has a long-lasting local anæsthetic action. We tentatively conclude that it may impair conduction in those neurones to which it gains access, and that its selectivity of action on adrenergic neurones may be related to its accumulation in these neurones.

The effects of 172*C*58 resemble those of bretylium but are more powerful. However, there are important qualitative differences. The adrenergic neurone-blocking action of 172*C*58 is considerably more rapid in onset and ends sooner, whether the drug is given by intravenous or subcutaneous injection. A related feature is its more rapid effect on isolated nerve-muscle preparations, from which, unlike bretylium, it may be readily removed by washing. Another major difference is that 172*C*58 caused no sympathomimetic effects in cats, except when several times the adrenergic neurone-blocking dose was given.

In two subjects given the drug intravenously and in two hypertensive patients given oral doses, 172*C*58 seemed a less active hypotensive agent than bretylium (Laurence, D. R., Moulton, R., and Rosenheim, M. L., personal communication). Its main value would seem to be as a pharmacological tool, its advantages over bretylium being its more rapid and more readily reversible action and its freedom from sympathomimetic properties.

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[1] Hey, P., and Willey, G. L., *Brit. J. Pharmacol.*, 9, 471 (1954).

[2] Exley, K. A., *Brit. J. Pharmacol.*, 12, 297 (1957).

[3] Taverner, D., cited by Bain, W. A., and Fielden, R., *Lancet*, ii, 472 (1957).

[4] Boura, A. L. A., Copp, F. C., and Green, A. F., *Nature*, 184, B.A.70 (1959).

[5] Boura, A. L. A., Green, A. F., McCoubrey, A., Laurence, D. R., Moulton, R., and Rosenheim, M. L., *Lancet*, ii, 17 (1959). Smirk, F. H., and Hodge, J. V., *ibid.*, ii, 673 (1959). Dollery, C. T., Emslie-Smith, D., and McMichael, J., *ibid.*, i, 296 (1960).

[6] Geus, R. J., McLean, R. A., Pasternack, J., Mattis, P. A., and Ullyot, G. E., *Fed. Proc.*, 18, 1, 1554 (1959).

[7] Boura, A. L. A., and Green, A. F., *Brit. J. Pharmacol.*, 14, 536 (1959).

Significance of Adipose Tissue Lipoprotein Lipase

THE post-heparin clearing factor in plasma has the same properties as tissue lipoprotein lipase; this is explained by Korn[1] by the fact that by the action of heparin lipoprotein lipase is released from the tissues in the blood stream. The greatest amount of lipoprotein lipase was found in adipose tissue[2]. A number of observations suggest indirectly the possible physiological significance of this enzyme in adipose tissue, particularly during the mobilization of fat from depots[3–7]. We have tried to elucidate the significance of adipose tissue lipoprotein lipase in rats by investigating the changes of its activity (*a*) after the administration of heparin and (*b*) during the mobilization and deposition of fat.

Immediately after killing the animal we removed the fat of the epididymis, froze it by means of liquid air, ground it to a fine powder from which we removed the fat in the cold, using acetone and ether, and afterwards dried it. The homogeneous powder was extracted with a solution of ammonia, *p*H 8·5, and the activity of lipoprotein lipase was estimated by determining the non-esterified fatty acids, using Dole's method[9], liberated during incubation with an activated artificial oil emulsion 'Ediol'[8] as a substrate (0·5 ml. extract: 2·5 ml. emulsion, temperature 37° C., incubation period 30 min.) and was expressed per 100 mgm. of dried tissue powder. 1 *M* sodium chloride, which is a specific inhibitor of lipoprotein lipase, inhibited esterolysis; this we consider evidence that in our experiments we actually estimate the enzyme.

The administration of heparin 'Novo' (500 μ/kgm. body-weight intraperitoneally), which causes a marked lipolytic activity of the blood, leads to a marked decrease ($P = 0{\cdot}05–0{\cdot}02$) of the lipoprotein lipase activity (Table 1, lines 1, 2). In a further series of experiments we determined the activity of lipoprotein lipase in adipose tissue of rats which had been fasted for 24 hr., and in similarly fasted rats with alloxan diabetes 2, 4 and 6 weeks after the administration of alloxan. No insulin was administered 48 hr. before the experiment. Under these conditions a marked mobilization of fat occurs. For comparison the activity in normal non-fasted animals and in animals which after 48 hr. fasting were re-fed for 20 hr. (Larsen mixture, glucose solution) was determined. In this case fat is not mobilized and in re-fed animals its deposition is much enhanced[10]. Table 1, lines 3 and 4, record the difference in the activity in rats fasting for 48 hr. and in re-fed rats. Fig. 1 summarizes the results of the entire series of experiments in relative figures. It is evident that under conditions when fat is mobilized the activity of lipoprotein lipase in adipose tissue is markedly reduced as compared with the activity in fed animals. Under the condition of an accentuated formation of depot fat in re-fed animals the activity is on the contrary markedly higher as compared with fed animals.

Table 1. RELEASE OF NON-ESTERIFIED FATTY ACIDS FROM THE SUBSTRATE

	No. of rats	No. of samples†	Microequivalent of non-esterified fatty acid ± S.E.	Statistical significance
1* Saline i.p.	15	5	29·02 ± 2 69	$P < 0{\cdot}05$
2* Heparin i.p. 500 μ/kgm.	15	5	21·88 ± 0 94	$P > 0{\cdot}02$
3 48 hr. fasting	18	6	6 32 ± 0·70	$P < 0{\cdot}001$
4 Refeeding	15	5	23·4 ± 3·19	

* Rats in experiments 1 and 2 had free access to food before being killed.

† Every sample was obtained by pooling adipose tissue from three animals.

We assume that in agreement with Korn's concept, which has been confirmed also by other authors[11,14], we can take the decrease of activity in the extract after the administration of heparin as a consequence of the liberation of lipoprotein lipase from adipose tissue. The high enzyme activity during fat deposition, contrary to the low activity during fat mobilization, is striking. The few papers published hitherto on the lipolytic activity of adipose tissue[12,13] do not reveal this fact. This can be explained by the use of a method which is not specific for the estimation of lipoprotein lipase[12]. Only Hollenberg's communication[14], which was published at a time when our work had been already completed and prepared for the press, has arrived at similar results though by a different approach.

As a result of our findings we may conclude, though it is only speculation so far, that during the formation

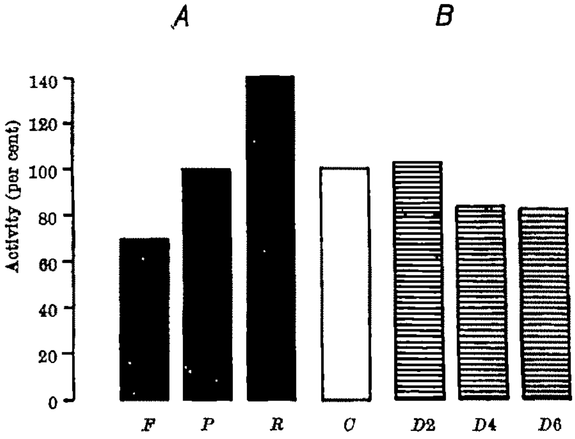

Fig. 1. Lipoprotein lipase activity of epididymal adipose tissue (*A*). *F*, decreased activity during fasting; *P*, 100 per cent activity in the post-absorptive state; *R*, increased activity after re-feeding. (*B*). *C*, 100 per cent activity of 24-h. fasted control rats; *D*2, *D*4, *D*6, diabetic rats, 2, 4 and 6 weeks, respectively, after administration of alloxan. Rats fasted 24 hr.

of fat in adipose tissue lipoprotein lipase is either synthesized or its activity is enhanced, which would suggest that this enzyme participates in the formation of fat depots. The finding of a reduced activity during fat mobilization must not be taken as evidence that lipoprotein lipase does not participate in this process. It is possible that during this process this enzyme is liberated by adipose tissue, just as after the administration of heparin—but much more slowly—to be transported to another site in the organism where its action is necessary.

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[1] Korn, E. D., *J. Biol. Chem.*, **215**, 1 (1955).
[2] Korn, E. D., and Quigley, jun., T., *Biochim. Biophys. Acta*, **18**, 143 (1955).
[3] Gordon, jun., R. S., *J. Clin. Invest.*, **36**, 810 (1957).
[4] Bragdon, J. H., and Havel, R. J., *Amer. J. Physiol.*, **177**, 128 (1954).
[5] Wenke, M., Wenkeová, J., Mühlbachová, E., and Škrobal, D., *Z. inn. Med.*, **13**, 104 (1958).
[6] Gordon, R. S., and Cherkes, A., *Proc. Soc. Exp. Biol. Med.*, **97**, 150 (1958).
[7] Wenkeová, J., and Páv, J., *Nature*, **184**, 1147 (1959).
[8] Zuckerman, L., and Singher, H. O., *Proc. Soc. Exp. Biol. Med.*, **89**, 315 (1955).
[9] Dole, V. P., *J. Clin. Invest.*, **35**, 150 (1956).
[10] Tepperman, J., and Tepperman, H. M., *Amer. J. Physiol.*, **193**, 55 (1958).
[11] Iselin, B., and Schuler, W., *Helv. Physiol. Acta*, **15**, 14 (1957).
[12] Renold, A. E., and Marble, A., *J. Biol. Chem.*, **185**, 367 (1950).
[13] Zemplényi, T., and Grafnetter, D., *Cas. lék. čes.*, **98**, 97 (1959).
[14] Hollenberg, C. H., *Amer. J. Physiol.*, **197**, 667 (1959).

Metabolism of Bemegride labelled with Carbon-14

Bemegride (β-ethyl β-methyl glutarimide), labelled with carbon-14 in both the α positions, is excreted largely in the urine and to a lesser extent in the bile and fæces when administered to mice, rats and guinea pigs[1]. Chromatograms of urine and bile from these animals were developed by the ascending technique using *n*-butanol/acetic acid/water (12 : 3 : 5, v/v). Autoradiographs of chromatograms of guinea pig bile showed only one spot (R_F 0·88) which was identical in mobility to the control-labelled bemegride. The urine of rats, collected 15–120 min. after administration of labelled bemegride, revealed two spots of R_F values 0·88–0·89 and 0·77–0·81. The faster moving fraction moved as one spot when labelled bemegride was added to the urine before chromatography. The spot with the lower mobility increased in intensity over the 15–120 min. period of urine collection. Urine from mice, 15–120 min. after bemegride injection, showed two spots with R_F values similar to those of rat urine, while urine collected 8 hr. after injection revealed a third spot of R_F 0·40–0·46. This spot became progressively more intense and the faster moving fractions correspondingly less up to 24 hr.

Bemegride labelled with carbon-14 was incubated with rat liver slices in Krebs–Ringer-phosphate solution for 5 hr. at 37° C. and two radioactive spots were revealed on chromatography of the liquid medium. The R_F values, 0·88 and 0·80, corresponded to those obtained from rat urine, suggesting that the liver is the site of bemegride metabolism *in vivo*.

Partition of rat urine with ether followed by chromatography of the two layers showed that the compound of R_F 0·80 was more soluble than bemegride in the aqueous phase (the concentrations were estimated as the relative density of the spots on the autoradiographs). This partition was not changed when determined at *p*H 1 or 7, which indicates that the metabolic fraction is of a similar nature to the unchanged glutarimide. The partition between mouse urine and chloroform revealed that while the spots of R_F 0·88 and 0·79 were distributed between the two layers in a manner similar to that between rat urine and ether, the fraction of R_F 0·45 was confined to the aqueous layer. This shows that the slowest moving fraction is extremely soluble in water.

Failure to detect carbon dioxide labelled with carbon-14 in the expired air of mice receiving labelled bemegride suggests that there is no drastic degradation of the glutarimide molecule. The metabolic products of bemegride detected in the urine of the mouse and rat do not appear to be hydrolysis products of this glutarimide, for the R_F values of β-ethyl β-methyl-glutaramic acid and -glutaric acid are 0·97 and 0·99 respectively in *n*-butanol/acetic acid/water. Succinimide has been shown to be hydrolysed to succinic acid *in vivo*[2] but this metabolic pathway does not appear to be available for the glutarimides. This is supported by the finding that the imide ring of α-ethyl α-phenyl glutarimide remains intact *in vivo*[3]. It is also interesting to note that *Lactobacillus* species are unable to hydrolyse α-amino-glutarimide[4]. β-(2-hydroxyethyl) β-methyl glutarimide, isolated from the urine of a patient who received a large dose of bemegride[5], was found to have an R_F value (0·80–0·83) similar to that of the slower-moving fraction in rat urine, but results from studies in rats involving unlabelled bemegride suggest that these two fractions are not identical.

In the chromatographic location of the hydroxy derivative of bemegride use was made of the technique of Rydon and Smith[6]. This method has proved a simple and valuable means of locating glutarimides on paper chromatograms. The limit of sensitivity for detection is 20–30 μgm. of glutarimide/cm.[2]. By using a 2 per cent starch 1 per cent potassium iodide solution a good definition of the spots is obtained.

It is clear that bemegride, while excreted partly unchanged, is metabolized *in vivo* to produce more water-soluble compounds which probably possess an intact glutarimide ring. The metabolic pathways in the rat and mouse are similar.

Work is in progress to isolate and identify these metabolic products from the urine of mice and rats receiving unlabelled bemegride.

I wish to thank Dr. L. F. Wiggins of Aspro-Nicholas, Ltd., Slough, for supplying bemegride and other glutarimide compounds and also Dr. N. E. W. McCallum for the sample of β-(2-hydroxyethyl) β-methyl glutarimide. I acknowledge receipt of a Nicholas Research Grant of the Welsh National School of Medicine.

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[1] Graham, J. D. P., and Nicholls, P. J., *Brit. J. Pharmacol.*, **14**, 35 (1959).
[2] Williams, R. T., "Detoxication Mechanisms" (Chapman and Hall, London, 1947).
[3] Sheppard, H., D'Assaro, B. S., and Plummer, A. J., *J. Amer. Pharm. Assoc., Sci. Ed.*, **45**, 681 (1956).
[4] Sondheimer, E., and Holley, R. W., *J. Amer. Chem. Soc.*, **79**, 3767 (1957).
[5] McCallum, N. E. W., *J. Pharm.* (*Lond.*), **7**, 276 (1956).
[6] Rydon, H. N., and Smith, P. W. G., *Nature*, **169**, 922 (1952).

Histochemical Evidence of Aminopeptidase Activity in Rat Pineal Gland

ALTHOUGH various functions have been suggested for the pineal gland[1], very few positive findings have been made concerning this gland[2,3]. In this communication histochemical evidence is given of a strong enzyme activity in the rat's pineal gland capable of splitting L-leucyl-β-naphthylamide hydrochloride. Recent studies[4,5] indicate that this activity is actually due to a group of peptidases, including 'leucine aminopeptidase'.

The pineal glands of about 20 albino rats were studied. After removal from the animals the glands were embedded in brain and rapidly frozen. Fresh frozen cold microtome sections were cut at 30μ. The sections were fixed on coverslips and incubated according to the method of Nachlas *et al.*[6], using diazotized *ortho*-dianisidine or diazonium salt of 4-amino-3 : 2′-dimethyl azobenzene as the coupling agents.

The pineal body of the rat exhibited a strong aminopeptidase activity (Fig. 1). The contrast between the staining of the brain tissue and the pineal tissue was very pronounced ; when the sections were incubated for 30 min. the brain tissue showed practically no staining at all, whereas the pineal tissue was intensely stained. The enzyme activity of the pineal body was not, however, quite as strong as it was in the rat kidney. The aminopeptidase activity seemed to be evenly distributed throughout the gland ; the choroid plexus was infrequently cut together with the pineal body, and it was found to exhibit a moderate enzyme activity.

Pineal bodies of rats of different age groups ranging from 1 to 16 months showed an equal aminopeptidase activity. No sex differences either could be noticed.

Peptidase activity was also tested using *ortho*-acetyl-5-bromoindoxyl as a substrate at an acid medium and diethyl-*p*-nitrophenylphosphate (*E*600) as an esterase inhibitor[7]. This reaction was positive in the rat's pineal body as well, demonstrating cathepsin-*C*-like peptidase activity.

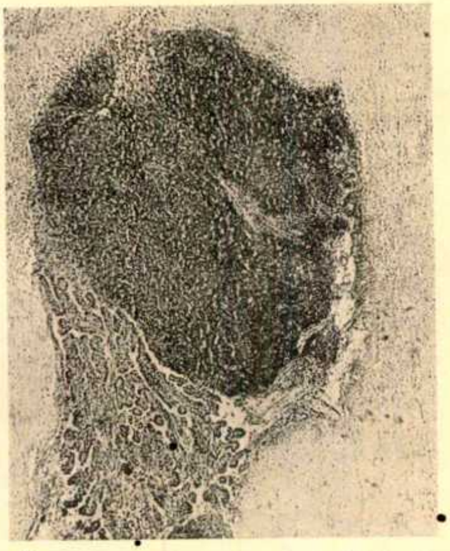

Fig. 1. Leucine aminopeptidase activity in pineal body of a normal adult rat. The choroid plexus beneath the gland shows enzyme activity as well, but the background staining of the brain tissue is due to decomposed diazonium salt (incubation time, 30 min.). (× *c.* 40)

So far as the proposed hormone production of the pineal gland is concerned, the present finding may be of interest. Pearse and Tremblay[8] have recently found an interesting correlation between the hormone production of the rat parathyroid gland and its content of histochemically demonstrable leucine aminopeptidase. The parathyroid hormone, on the other hand, is known to be a mixture of polypeptides. The existence of aminopeptidase activity can be taken to suggest secretion of some agent of a protein nature by the rat pineal gland.

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[1] Kitay, J. I., and Altschule, M. D., "The Pineal Gland" (Harvard University Press, 1954).

[2] Leduc, E. H., and Wislocki, G. B., *J. Comp. Neur.*, **97**, 241 (1952).

[3] Giarman, N. J., Day, S. M., and Pepeu, G., *Fed. Proc.*, **18**, 394 (1959).

[4] Waldschmidt-Leitz, E., and Keller, L., *Hoppe-Seyl. Z.*, **309**, 228 (1957).

[5] Glenner, G. G., Burstone, M. S., and Meyer, D. B., *J. Nat. Cancer Inst.*, **23**, 857 (1959).

[6] Nachlas, M. M., Crawford, D. T., and Seligman, A. M., *J. Histochem. Cytochem.*, **5**. 264 (1956).

[7] Hess, R., and Pearse, A. G. E., *Brit. J. Exp. Path.*, **39**, 292 (1958).

[8] Pearse, A. G. E., and Tremblay, G., *Nature*, **181**, 1532 (1958).

Clearances of Endogenous α-Ketoglutaric and Pyruvic Acid in Man

THE clearances of endogenous α-ketoglutaric and pyruvic acids were investigated in six healthy persons and in ten patients with damaged kidney parenchyma. All persons were confined to bed during the whole experiment. α-Keto-acids were determined after their conversion to corresponding hydrazones which were separated by paper electrophoresis[1].

In all patients with kidney damage the creatinine clearance was lowered. The urine was collected in eight 2-hr. portions during a day and in two 4-hr. portions in the course of a night. (The mean values of clearances of α-ketoglutaric acid and of pyruvic acid was calculated from ten collection periods.)[2] The blood was deproteinized immediately after venipuncture and the clear supernatant was processed as usual.

The values of both the α-keto-acids in blood varied during a 24-hr. period. The variations averaged ± 15 per cent in the case of α-ketoglutaric acid and ± 20 per cent of values of pyruvic acid. 1·00–2·70 mgm. of α-ketoglutaric acid per 100 ml., and 0·90–1·70 mgm. of pyruvic acid per 100 ml. of urine were usually found. (The concentration was found to be dependent upon diuresis.) Women excreted more of these acids during 24 hr. than men ; this difference being accentuated in the case of α-ketoglutaric acid (Table 1).

The mean values of clearances of endogenous α-ketoglutaric and pyruvic acids were higher in women as compared with men (Table 1). In patients with kidney damage the values of both clearances were lowered ; the lowest one was found in nephritis of middle degree.

Highest values of clearances were observed between 1 p.m. and 5 p.m., and between 7 p.m. and 9 p.m.

The cause of different concentrations of both α-keto-acids in blood and urine is to be seen in kidneys. The kidney does not only eliminate the α-keto-acids, it participates in their metabolism, too[3]. The transaminations, oxidative deaminations, and oxidative

Table 1. MEAN VALUES OF α-KETOGLUTARIC AND PYRUVIC ACIDS IN BLOOD AND URINE DURING 24 HR. AND THE CLEARANCES IN HEALTHY PERSONS, AND IN PATIENTS WITH KIDNEY DAMAGE

		No. of cases	Corresponding keto-acid	Mgm. per 100 ml. of blood	Mgm. in 24 hr. urine	Mean clearance values in 24 hr. (ml. per min.)
Healthy	women	3	*KG*	0·14 ± 0·05	18·66 ± 4·41	8·47 ± 1·86
			P	0·44 ± 0·09	11·41 ± 3·10	1·76 ± 0·43
	men	3	*KG*	0·15 ± 0·06	11·95 ± 2·52	7·67 ± 1·91
			P	0·51 ± 0·12	9·62 ± 2·67	1·46 ± 0·42
Kidney damage	women	5	*KG*	0·16 ± 0·07	17·36 ± 3·81	7·93 ± 1·66
			P	0·58 ± 0·13	11·36 ± 2·95	1·52 ± 0·39
	men	5	*KG*	0·17 ± 0·06	11·99 ± 2·16	5·80 ± 1·10
			P	0·65 ± 0·16	10·27 ± 2·15	1·30 ± 0·36

KG, α-Ketoglutaric acid ; *P*, pyruvic acid,

decarboxylations will be of principal importance here. It is possible to explain the relatively high concentration of α-ketoglutaric acid in urine (almost 10 times higher than that in blood) by either its lower reabsorption in renal tubules or by its higher elimination rate in the kidneys.

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[1] Zelníček, E., *Nature*, **184**, 727 (1959).

[2] Zelníček, E., *Cas. lék. ces.* (in the press).

[3] Nordmann, J., and Nordmann, R., *Clin. Chem.*, **3**, Supp. 462 (1957).

Respiratory Function of the Swim-bladder of *Notopterus* (Lacepede)

BRIDGE[1], working on the swim-bladder of *Notopterus borneensis* (Bleeker), and Srivastava[2], on that of *Notopterus chitala* (Buchanan-Hamilton), concentrated their attention only on the general plan of the organ.

A detailed account of the swim-bladder and its relation with the internal ear in the genus *Notopterus* has already been published by me[3]. During the course of study, the fish lived for an indefinite period in the laboratory even when the water in the aquarium became foul and contained less than 0·5 p.p.m. of dissolved oxygen for most of the time. The specimens of *N. chitala* and *N. notopterus* (Pallas) kept in the aquarium rose to the surface to gulp air at regular intervals. If they were prevented from rising they died in 4–6 hr. The duration of time between the two consecutive gulps of air varied generally from 2 to 8 min. under normal conditions. The frequency of surface visits increased after air was removed from the swim-bladder forcibly by pressing at its caudal cæcum (Fig. 1) or by drawing air out of the organ by means of a hypodermic syringe. Thus it may be presumed that a part of the air gulped by the fish enters its swim-bladder through its pneumatic duct at intervals and thus brings about a regular exchange of gases between the cavity of the organ and the exterior.

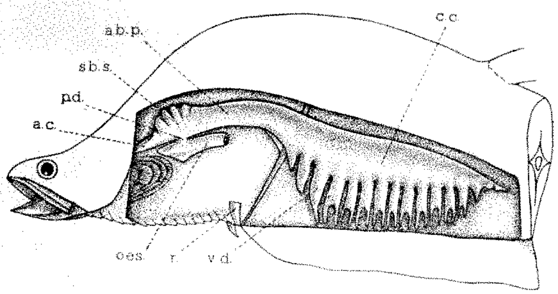

Fig. 1. Lateral view of swim-bladder of *Notopterus chitala*. *ab.p.*, Abdominal portion ; *a.c.*, auditory cæcum ; *c.c.*, caudal cæcum ; *oes.*, œsophagus ; *p.d.*, pneumatic duct ; *r.*, rectum ; *sb.s.*, subspherical sac ; *v.d.*, ventral diverticulum

Histological study of the swim-bladder wall shows that the tunica interna is abundantly supplied with blood capillaries. The blood is brought in close contact with the contained gases which diffuse through the thin layer of epithelial cells. The circulation of blood is modified in the organ to facilitate its function as an accessory respiratory organ on the plan observed by Carter and Beadle[4]. The blood supply is from the dorsal aorta and the oxygenated blood is drained back to the heart through the posterior cardinal vein.

Further evidence of aerial respiration came from the analyses of gases from the swim-bladder of the fish. The gases were drawn from the caudal cæcum of the swim-bladder by means of a hypodermic syringe and analysed by an apparatus made in the laboratory on the plan described by Fry[5]. Immediately after *N. chitala* had gulped air, the percentages of oxygen and carbon dioxide in the gases of its swim-bladder were found to be (mean) 16 and 0·5 respectively. But when the fish were prevented from gulping air for 10 min., the percentages of oxygen and carbon dioxide in the gases of its swim-bladder were found to be (mean) 6 and 4 respectively. The water contained 1 p.p.m. of dissolved oxygen and 20 p.p.m. of free carbon dioxide during the above observations.

The air-breathing habit of *Notopterus* has not been recorded so far. Behaviour of *Notopterus* regarding its aerial and aquatic respiration under various free carbon dioxide and dissolved oxygen contents in water is being studied. Work on the physiological aspects of its aerial respiration is in progress, and a detailed account will be published elsewhere.

I am grateful to Dr. M. Chandy, under whose guidance the above work was done. Thanks are due to Prof. M. L. Bhatia, for encouragement and facilities offered in the Department.

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[1] Bridge, T. W., *J. Linn. Soc.* (*Zool.*), **27**, 503 (1899–1900).

[2] Srivastava, P. N., *Proc. Nat. Acad. Sci.* (*India*), **25**, B, III–IV, 46 (1955).

[3] Dehadrai, P. V., *J. Zool. Soc. India*, **9** (1), 50 (1957).

[4] Carter, G. S., and Beadle, L. C., *J. Linn. Soc.* (*Zool.*), **37**, 205 (1931).

[5] Fry, F. E. J., *Canad. J. Res.*, E, **27**, 188 (1949).

HÆMATOLOGY

Significance of Thrombin-*E* in the Prothrombin Activation Sequence

THROMBIN-*E*, or esterase thrombin, is an enzyme that uses fibrin monomer and fibrin as a substrate, but not fibrinogen[1,2]. Blood does not clot in the presence of thrombin-*E*, but on the contrary there is a tendency for lysis. It is derived from prothrombin, and arises before thrombin-*C*, which is the traditional enzyme considered in the blood coagulation literature[3]. In this report we emphasize the significance of thrombin-*E* in a prothrombin activation sequence. We describe some pharmacological effects of a purified thrombin-*E* preparation, chief of which is its fibrinolytic effect upon intravenous infusion in dogs. This *in vivo* fibrinolysis is far more powerful than its *in vitro* fibrinolytic effect. These facts are consistent with previous postulations[4,5] as follows: certain chemo-receptors excite themselves with thrombin and through a reflex arc call into play a neuro-humoral mechanism involving a physiological anti-coagulating system.

Two versions of the prothrombin activation sequence have been presented, but with no greater completeness than was comprehensible in view of the available information at the time[6,7]. With the knowledge that thrombin-*E* arises before thrombin-*C* an expanded sequence can be considered. First, there is prothrombin, which is recognized as a distinct single entity on the basis of isolation work[8,9]. It may activate according to the following sequence:

Prothrombin	becomes thrombin in two-stage analytical reagents
↓	
Prothrombin derivative I	does not become thrombin in two-stage analytical reagents
↓	
Prothrombin derivative II	becomes thrombin in two-stage analytical reagents
↓	
Thrombin-*E*	uses fibrin monomer and fibrin as substrate, but not fibrinogen
↓	
Thrombin-*C*	uses fibrinogen, fibrin monomer and fibrin as a substrate
↓	
Thrombin-*E*	uses fibrin monomer and fibrin as substrate, but not fibrinogen

Thrombin-*E* can be obtained from prothrombin in several ways. For example, by activating prothrombin in calcium solutions[2], or by letting thrombin-*C* preparations deteriorate[1]. But these ways are not practicable, for some thrombin-*C* usually remains and the yields are low. Later we hope to report on the derivation of a type of thrombin-*E* obtained by activation of acetylated prothrombin. It seems to be as convenient a way as by acetylation of thrombin-*C* [10], which was the method employed to get the material for our pharmacological studies.

Having achieved the preparation of homogeneous (ultracentrifuge) thrombin-*E* [10] we were ready to see, for the first time, some of its pharmacological effects. Approximately 15 mgm., dissolved in 100 ml. physiological saline solution, were infused in the femoral vein of a dog previously given 'Nembutal' anæsthesia. The infusion lasted over a period of an hour. There were no changes in blood pressure, respiratory and cardiac rate, or temperature. At completion of the infusion blood was drawn. Sometimes it clotted and then lysed. Sometimes the fibrinogen was depleted and there was no clotting. Of the several blood analyses done it was of interest to see a decrease in platelet and white cell count, and almost a two-fold increase in blood sugar and cholesterol concentration. By the end of 24 hr. all blood values were back to normal. The profound fibrinolysis is due to physiological mechanisms activated by thrombin-*E*. The equivalent concentrations or even a hundred times more of thrombin-*E* added to dog blood in test-tubes was never followed by visible lysis. On the other hand, plasma from dogs given infusions could be mixed with a standard fibrin clot and lysis was easily measured.

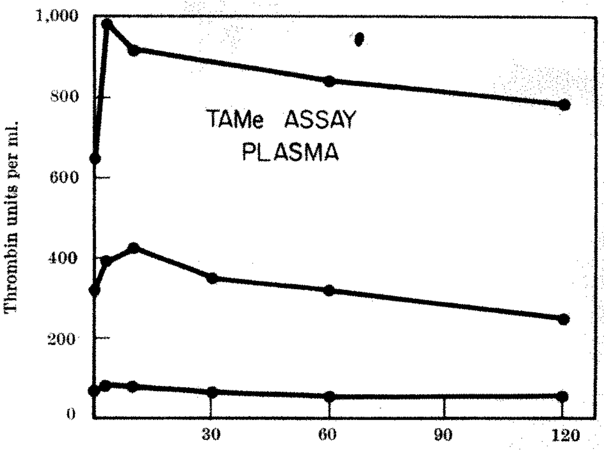

Fig. 1. Acetylated thrombin, in concentrations of 1,300, 650 and 130 units per ml., was mixed with an equal volume of defibrinated cow plasma. The activity at first increased and then declined to lower levels. The initial increase in activity was proportional to the thrombin-*E* concentration

To clarify the question of thrombin-*E* neutralization by antithrombin we made several analyses. In contrast to the rapid neutralization of the activity in thrombin-*C* [11] there seems to be an intensification of esterase activity during the first half hour after plasma is mixed with thrombin-*E* (Fig. 1). If the concentration of the latter is not too high it eventually is neutralized by antithrombin. Thus it is quite possible to have thrombin-*E* activity in the face of plasma antithrombin.

There are two facts that can be used to develop a perspective on the significance of thrombin-*E* in the prothrombin activation sequence. First, it arises before thrombin-*C*, and secondly, it activates fibrinolytic mechanisms in the intact mammal. The control of prothrombin activation is by a group of anticoagulants and procoagulants functioning in dynamic equilibrium[12]. With procoagulants gaining ascendency, thrombin-*E* can form and excite physiological mechanisms concerned with fibrinolysis. Even excess of thrombin-*E* is without danger so far as fibrinogen activation is concerned, and it is also not a catalyst for prothrombin activation[2]. However, if the prothrombin activation sequence goes farther we obtain thrombin-*C*. It can also excite fibrinolytic mechanisms[4,5], presumably through use of its thrombin-*E* component, but in 'excess' concentration it activates fibrinogen and clotting follows. Our results also account for the observed therapeutic effect obtained in cases of thrombosis where small amounts of crude thrombin-*E* preparations were infused[13]. The view that chemo-receptors react to the appearance of thrombin[4,5] focuses attention on events at an advanced stage of prothrombin activation. Already before that, thrombin-*E* can be effective, and it probably is constantly functioning to help maintain the fluidity of the blood, for it could be present in significant concentration at all times. For example, we made a few analyses and found five units of esterase activity in dog plasma and thirteen units in 2-hr.-old dog serum. This may not have been due to thrombin, but the same method for analysis was used as is routinely employed for the measurement of thrombin-*E* activity[1,2].

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[1] Landaburu, R. H., and Seegers, W. H., *Proc. Soc. Exp. Biol. Med.*, **94**, 708 (1957).

[2] Seegers, W. H., and Landaburu, R. H., *Amer. J. Physiol.*, **191**, 167 (1957).

[3] Landaburu, R. H., and Seegers, W. H., *Amer. J. Physiol.*, **197**, 1178 (1959).

[4] Kudrjashov, B. A., and Ulytina, P. D., *Nature*, **182**, 397 (1958).

[5] Kudrjashov, B. A., *Nature*, **184**, 454 (1959).

[6] Seegers, W. H., and McClaughry, R. I., *Proc. Soc. Exp. Biol. Med.* **72**, 247 (1949).

[7] Seegers, W. H., Casillas, G., Shepard, R. S., Thomas, W. R., and Halick, P., *Canad. J. Biochem. Physiol.*, **37**, 775 (1959).

[8] Seegers, W. H., *Record Chem. Prog.*, **13**, 143 (1952).

[9] Miller, K. D., *J. Biol. Chem.*, **231**, 987 (1958).

[10] Landaburu, R. H., and Seegers, W. H., *Canad. J. Biochem. Physiol.*, **37**, 1361 (1959).

[11] Seegers, W. H., Miller, K. D., Andrews, E. B., and Murphy, R. C., *Amer. J. Physiol.*, **169**, 700 (1952).

[12] Landaburu, R. H., and Seegers, W. H., *Amer. J. Physiol.*, **193**, 169 (1958).

[13] Brambel, C. E., *Ann. N.Y. Acad. Sci.*, **68**, 67 (1957).

Erythrocyte Glucose-6-Phosphate Dehydrogenase-deficient Subjects: Enzyme-Level in Saliva

AN erythrocyte glucose-6-phosphate dehydrogenase deficiency has been described among Negroes, non-Ashkenazi Jews and Italians, mostly of Sardinian origin[1-3]. This enzymatic abnormality is known to be associated with an increased susceptibility to hæmolysis following ingestion of a number of drugs as well as of *fava* beans[4,5], and appears to be transmitted as a sex-linked incompletely dominant trait[6].

Recently, a low enzyme-level in the platelets, white blood-cells and liver-homogenates of affected subjects was demonstrated in our laboratory[7,8]. Marks *et al.* have also found low enzyme-levels in leucocytes of affected Caucasians in contrast to their previous reports on normal glucose-6-phosphate dehydrogenase activity in leucocytes of affected Negroes[9]. This apparent difference, in itself noteworthy, may be due to either (*a*) at least two or more independent genetic mutations, or (*b*) an effect of different genetic modifiers, which determine the degree of the enzyme defect in the population groups studied.

Since this genetic abnormality affects many primitive peoples, from whom we have had difficulties in obtaining blood, the search for other sampling methods to investigate this defect was undertaken. Saliva was found to be suitable for this purpose. Samples of it were obtained from twenty-six fasting subjects (controls), and from fourteen 'test cases', all males, deficient in glucose-6-phosphate dehydrogenase. The saliva samples were spun down at 1,000*g* for 15 min., 1 ml. of the supernatant being used for the assay. Glucose-6-phosphate dehydrogenase and 6-phosphogluconic dehydrogenase were assayed by previously described methods[8,10,11]. The enzymatic activity is expressed in units per mgm. of protein. The phosphogluconic dehydrogenase activity in enzyme-deficient individuals was not significantly different from that of normal subjects.

Table 1. GLUCOSE-6-PHOSPHATE DEHYDROGENASE (G-6-P.D.) ACTIVITY IN SALIVA OF NORMAL AND ENZYME-DEFICIENT SUBJECTS

Subjects	Number	G-6-P.D. level in units*	
		Range	Mean
Control	26	2–27	$8 \pm 6{\cdot}3$
Sensitive	14	0–1·22	$0{\cdot}54 \pm 0{\cdot}4$

$$^{*}\text{Unit} = \frac{(OD/\text{min.})}{(\text{mgm. protein})} \times 10^{3}$$

As can be seen in Table 1, the level of glucose-6-phosphate dehydrogenase in the saliva of 'test cases' is significantly lower than that in normal controls, ($P > 0{\cdot}001$) thus demonstrating that saliva, which is easily obtained, is suitable for the detection of enzyme-deficient subjects.

Preliminary investigation of the skin of normal and enzyme-deficient subjects has revealed that low enzyme-levels of the latter group are also demonstrable. These findings further support our observation that the genetic enzymatic defect in our population affects all tissues, not only the blood cells.

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[1] Carson, P. E., Flanagan, C. L., Ickes, C. E., and Alving, A. S., *Science*, **124**, 484 (1956).

[2] Szeinberg, A., Sheba, C., and Adam, A., *Nature*, **181**, 1256 (1958).

[3] Beutler, E., Dern, R. J., Flanagan, C. L., and Alving, A. S., *J. Lab. Clin. Med.*, **45**, 286 (1955).

[4] Szeinberg, A., Sheba, C., Hirshorn, N., and Bodonyi, E., *Blood*, **12**, 603 (1957).

[5] Sansone, G., and Segni, G., *Boll. Soc. Ital. Biol. Sper.*, **32**, 456 (1956).

[6] Childs, B., Zinkham, W., Browne, E. A., Kimbro, E. L., and Torbert, J. V., *Bull. Johns Hopkins Hosp.*, **102**, 21 (1958).

[7] Ramot, B., Szeinberg, A., Adam, A., Sheba, C., and Gafni, D., *J. Clin. Invest.*, **38**, 1659 (1959).

[8] Ramot, B., Fisher, S., Szeinberg, A., Adam, A., Sheba, C., and Gafni, D., *J. Clin. Invest.*, (in the press).

[9] Marks, P. A., *et al.*, *Biochem. Biophys. Res. Comm.*, **1**, 199 (1959).

[10] Kornberg, A., and Horecker, B. L., "Methods in Enzymology", edit. by Colowick, S. P., and Kaplan, N. O., **1**, 323 (Academic Press, Inc., New York, 1955).

[11] Horecker, B. L., and Sayrniotis, P. Z., *J. Biol. Chem.*, **193**, 371 (1951).

Inheritance of a New Group-Specific System demonstrated in Normal Human Sera by means of an Immuno-Electrophoretic Technique

IN a previous communication[1] a new group-specific system was demonstrated by means of a modified immuno-electrophoretic technique[1-3], and it has been shown that this system is not related to the hapto-globin groups[1,2]. Thus, by the content of either a fast-moving precipitating component (G.c.1), a slow-moving precipitating component (G.c.2) (Fig. 1), or a precipitating component which gives an immuno-precipitate with two peaks, human sera can be classified into three qualitatively different groups, which have been called groups 1, 3 and 2 respectively.

It has been shown that the precipitate with two peaks, or in less optimal circumstances, a plateau-like shape, can also be obtained by mixing sera belonging to groups 1 and 3 in equal amounts and subjecting them to immuno-electrophoresis. In all these instances (7 experiments with different sera) a precipitate is obtained which is indistinguishable from the precipitating component with two peaks found in certain normal sera, which are thereby characterized as belonging to serum group 2, and from these experiments presumably contain both the fast-moving and slow-moving precipitating components (Fig. 2)[1].

These group-specific components differ from the haptoglobins by their precipitates not taking the benzidine-stain after addition of hæmoglobin, they do not show any change in electrophoretic position after addition of hæmoglobin and are further found in all human sera hitherto investigated, including a few sera which were found to lack any haptoglobin demonstrable by starch-gel and agar-gel electrophoresis[3]. The group-specific components are found in the α_2-region occupying electrophoretic positions approximately corresponding to the haptoglobins 1–1 and 2–2, when sera are investigated without addition of hæmoglobin; but they are not correlated to the haptoglobin-groups determined in 30 sera by means of starch-gel electrophoresis.

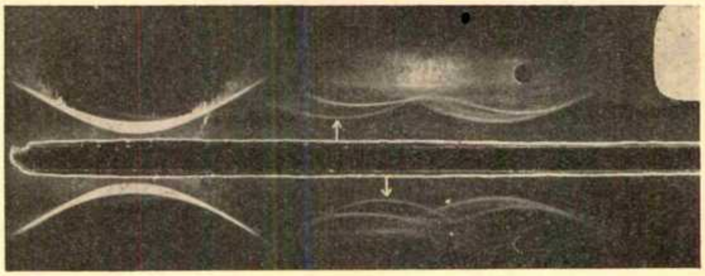

Fig. 1. Immuno-electrophoretic pattern of two normal sera characterized as belonging to group 1 (upper basin) and group 3 (lower basin). The group-specific components are indicated by arrows. To the far left, the position of albumin is shown

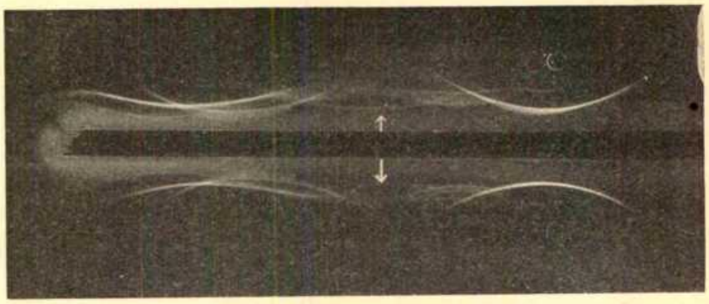

Fig. 2. Immuno-electrophoretic pattern obtained by mixing equal amounts of the same sera as in Fig. 1 (upper basin) as compared with a normal serum (lower basin) containing a precipitate with two peaks or at least a plateau-like form, the serum therefore characterized as belonging to group 2 or the heterozygote. A different immune serum is used from that in Fig. 1. This does not, however, affect the determination of the group-specific components

While it has not yet been excluded that these group-specific components might be identical with, for example, the post-albumins[4], the above-mentioned preliminary denotations will still be used.

In the present communication the results of a genetical study of mother–child material (kindly obtained from Prof. Birger Broman, State Institute for Blood Group Serology, Stockholm) and family material (kindly obtained from Prof. Rune Grubb, Bacteriological Department, Lund) will be given.

Family data are summarized in Table 1. They are in agreement with the interpretation that the character is monogenetically inherited and that types 1 and 3 represent the two homozygous classes and type 2 is the heterozygote.

Table 1

Parental phenotypes		No. of families	No. of children belonging to type			Total
Father	Mother		1	2	3	
1	1	6	30	0	0	30
1	2	2	4	2	0	6
2	1	4	4	7	0	11
2	2	3	2	3	1	6
1	3	1	0	2	0	2
3	1	1	0	3	0	3

Table 2

	Number of individuals belonging to type			Total
	1	2	3	
Observed	161	108	23	292
Expected	158·3	113·4	20·3	292

$\chi^2 = 0\cdot662$, 1 degree of freedom. $P = 0\cdot3$–$0\cdot5$.

A randomly sampled specimen of mother–child material confirmed this hypothesis, in that among 76 mothers of type 1 no one had a child of type 3, and among 14 mothers of type 3 no one had a child of type 1. Gene frequencies were estimated from these data, tabulated in Table 2, using the scoring method devised by Cotterman[5] for material composed of related individuals. The frequency of the allele responsible for the fast-moving component was found to be 0·74 and the frequency of the other allele 0·26. A χ^2-test for the agreement with the postulated manner of inheritance and for genetic equilibrium was performed (Table 2). The deviations from the expected frequencies are very small and not statistically significant.

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[1] Hirschfeld, J. K.-E., *Acta Path. et Microbiol. Scand.*, **47**, 160 (1959).

[2] Hirschfeld, J. K.-E., *Acta Path. et Microbiol. Scand.*, **46**, 229 (1959).

[3] Hirschfeld, J. K.-E., *Acta Path. et Microbiol. Scand.*, **47**, 169 (1959).

[4] Smithies, O., *Biochem. J.*, **71**, 585 (1959).

[5] Cotterman, C. W., in Kempthorne, Bancroft, Gowen, and Lush "Statistics and Mathematics in Biology" (Iowa State College Press, 1954).

PLANT PHYSIOLOGY

Effect of Coumarin on the Growth of *Rhodotorula rosea* Cultures

Coumarin ($C_6H_4OCOH{=}CH$) and closely related compounds are strong regulators of the germination and growth of seedlings of different species of plants[1]; the stimulatory[2] or the inhibitory[3,4] effect depends on the concentration of coumarin and on the species of plant tested. Unfortunately, there is almost no information concerning the effect of coumarin on the growth of micro-organisms[5] and we have, therefore, carried out tests with *Rhodotorula rosea*.

Into 100 ml. Erlenmeyer flasks were poured 24·5 ml. synthetic nutrient medium[6] (twice condensed), 24·5 ml. special solution of coumarin, or distilled water in the case of control, and sterilized in an autoclave. After cooling, 1 ml. suspension of *Rh. rosea* (48-hr. culture grown on sweet beer, by dilution 1 : 10,000) was added to each flask, which was then placed in a thermostat at a temperature of 27° C.

The growth of micro-organisms was measured by microscopic observations of preparations stained with methylene blue solution according to Fink and Kühles[7] in Thoma's counting chamber. Besides the whole number of cells, dead and budding cells were also counted.

The results are presented in Tables 1 and 2.

Table 1. Effect of Coumarin Concentrations on Rate of Growth of *Rhodotorula rosea* after Various Times

Concentration of coumarin (p.p.m.)	Percentage of growth after (hr.) 48	72	96
0 (control)	100	100	100
1·0	81·2	72·4	64·0
10·0	152·9	112·3	78·4
100·0	77·1	53·8	36·1
1,000·0	6·4	4·6	2·0

Table 2. Effect of Coumarin on the Degree of Budding and Dead Cells

Concentration of coumarin (p.p.m.)	Percentage budding after (hr.)			Percentage dead after (hr.)		
	48	72	96	48	72	96
0 (control)	72·6	67·8	77·1	8·3	6·2	4·7
1·0	68·0	69·9	73·9	8·6	9·4	6·4
10·0	78·7	81·0	78·2	5·4	5·3	6·5
100·0	55·6	57·9	68·8	9·5	20·3	14·7
1,000·0	14·4	29·4	0	42·8	58·9	96·1

Note: all numbers are the average of 3–4 repetitions.

The results in Table 1 show that coumarin can stimulate or inhibit the growth of culture *Rh. rosea*, depending on time and concentration. At a concentration of 1,000 p.p.m. it almost totally inhibits development of the yeast: in a 48-hr. culture there are only 6 per cent of cells compared with the control; in this case 43 per cent of cells are dead. In a 96-hr. culture there are only 2 per cent, and in this case almost all cells are dead. The fact must be recognized that the cells alive in a culture such as this change very much morphologically (unpublished work).

Interesting results are obtained in cultures after adding coumarin at a concentration of 10 p.p.m. After 48 hr. in such a culture there is an increase of the whole amount of cells to the extent of 50 per cent (152–100 per cent of control); in cultures lasting 72 hr. stimulation decreases to about 12 per cent. In cultures lasting 96 hr., however, instead of stimulation, an inhibition of about 20 per cent of growth is observed. It is important that the decrease of stimulation with ensuing inhibition is proportional to the time of growing culture on nutrient medium with a supply of coumarin.

Parallel to the increase of the whole amount of cells in cultures treated with coumarin at a concentration of 10 p.p.m. there is an increase in the number of budding cells, but there is a decrease in the number of dead cells (expressed as a percentage of the whole number of cells in the given series taken as 100 per cent); by passing from stimulation to inhibition, at the same time, the increase of the amount of dead percentage of cells is observed.

An important problem is that coumarin in a concentration of 1 p.p.m. shows inhibitory action.

On the basis of the investigations described above it may be suggested that coumarin has a decisive influence on the living process of the yeasts (we obtained similar results on *Saccharomyces cerevisiae*), and it is very probable that it influences the development of other strains of micro-organisms.

We thank Prof. W. Moycho for his critical remarks made while working on this problem.

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[1] Blaim, K., *Kosmos* (Warsaw), Ser. A, Biol., **6**, 243 (1957).

[2] Neumann, J., *Science*, **129**, 1675 (1959).

[3] Ishikawa, S., *Kumamoto, J. Sci.*, **2**, 97 (1955).

[4] Misra, G., and Patnaik, S. N., *Nature*, **183**, 989 (1959).

[5] Moewus, F., and Banerjee, B., *Nature*, **168**, 561 (1951).

[6] Fiodorow, M., Ćwiczenia praktyczne z mikrobiologii,, 83 (PWRiL, Warszawa, 1952).

[7] Jørgensen, A., trans. by Hansen, A., "Micro-organisms and Fermentation", 535 (Charles Griffin and Co., Ltd., London, 1948).

A Reversible Inhibition of Photosynthesis in Synchronized Cultures of Algae

Steady-state algal suspensions have been a favourite subject in studies of inhibitory effects of light on photosynthesis[1–5]. Light intensities several times higher than the intensity of full sunlight were often employed. The unfavourable effects of strong light were seen in the decreased photosynthetic rates[1–4] and/or in the destruction of the photosynthetic pigments[5]. A recovery process has been observed after transferring the organism into dark or to lower light intensities[3,4,5]. An emphasis on the dependence of the photo-inhibition on a very strong light has obscured the much wider nature of the phenomenon. The general employment of steady-state suspensions precluded an insight into the complexity of the situation on the cellular level.

A steady-state microbial population consists of cells of different developmental stages. Any characteristic obtained for a steady-state suspension is a statistical average from the characteristics of the individual cells and, like any average, it may or may not be intrinsic to any portion, or, strictly speaking, even to a single cell, of a given population. It has been shown that in the course of the life-cycle *Chlorella* cells pass through stages which differ in the levels of metabolic and growth activity[6–9], in the contents of cell constituents[10,11], and in the sensitivity to environmental factors[12].

The effect of light on photosynthesis in cells of different developmental stages also proved to be

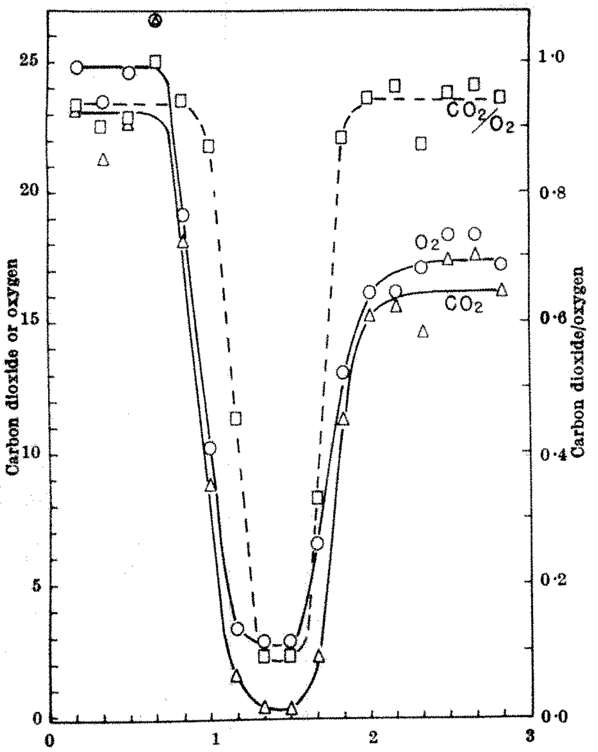

Fig. 1. Time course of the gas exchange during a photosynthetic experiment in 3-hr.-old cells of *Chlorella* 7–11–05. Carbon dioxide consumption and oxygen evolution in mm.^{3} of gas per mm.^{3} of cells per 10-min. intervals. Assimilatory quotients (carbon dioxide/oxygen) for the corresponding intervals. Note recovery of rates for all measurements

different[13]. Cells of a certain intermediate age are characterized by a higher position of the light-saturating point and by higher rates of photosynthesis at light saturation. In the course of a photosynthetic experiment these cells show a peculiar phenomenon. The rate of the gas exchange first declines; then, after reaching a low point, it increases and may eventually reach or surpass the level of the initial rate at the start of the experiment.

The results depicted in Fig. 1 were obtained with a synchronized suspension of the high-temperature strain, *Chlorella* 7–11–05[7,8]. Cells 3 hr. old were used. Studies were done with the two-vessel technique in 0·02 *M* potassium dihydrogen phosphate-suspending fluid at *p*H 4·5. Temperature during both the culturing period and photosynthesis measurements was maintained at 39° C. Illumination during photosynthesis studies was provided by incandescent lamps with the intensity at the level of the reaction vessels near 2,000 foot candles. This intensity is slightly higher than the saturating light intensity for the steady-state suspensions of this strain (near 1,600 foot candles)[14].

As seen in Fig. 1 both the oxygen evolution and carbon dioxide consumption undergo sharp declines with time. Carbon dioxide consumption is affected more than oxygen evolution and, consequently, the assimilatory quotient (carbon dioxide/oxygen) drops to a very low level. In some experiments the carbon dioxide consumption was even observed to turn into carbon dioxide evolution and the oxygen evolution into oxygen consumption.

After reaching a low level the rates of gas exchange start to rise. Eventually they may reach the initial level or even surpass it. The rate of the recovery of carbon dioxide consumption is higher than that of oxygen evolution and, as a result, the assimilatory quotient gradually rises to the initial level at the start of the experiment.

For comparison, the time course of the events in a steady-state suspension of the same strain and under similar conditions is depicted in Fig. 2. The rate of oxygen evolution declines with the time in accordance with earlier observations by Emerson[1]. Carbon dioxide consumption also shows a gradual though slightly greater decrease with time. As the result, assimilatory quotient slightly decreases during the course of the experiment. Changes in the rates of the gas exchange in the steady-state suspension are not nearly so dramatic as in synchronized 3-hr. cells and recovery of the rate does not occur.

A clue to the discrepancy in the time course of photosynthetic gas exchange in synchronized 3-hr.-old cells (Fig. 1) and in a steady-state suspension (Fig. 2) is provided by an understanding of the diversity of cellular characteristics among members of a steady-state microbial population. It will be shown in following publications that algal cells of later developmental stages have a very low if any capacity for a recovery of photosynthetic rate in the course of the experiment. The time course of the photosynthetic gas exchange in older cells would be described by a constantly decreasing rate with the slope of the curve much steeper than that depicted in Fig. 2 for the steady-state suspension. At any given time during the measurement of photosynthesis the rate of the gas exchange for the steady-state suspension is an average of the rates of cells in all possible developmental stages. The preponderance

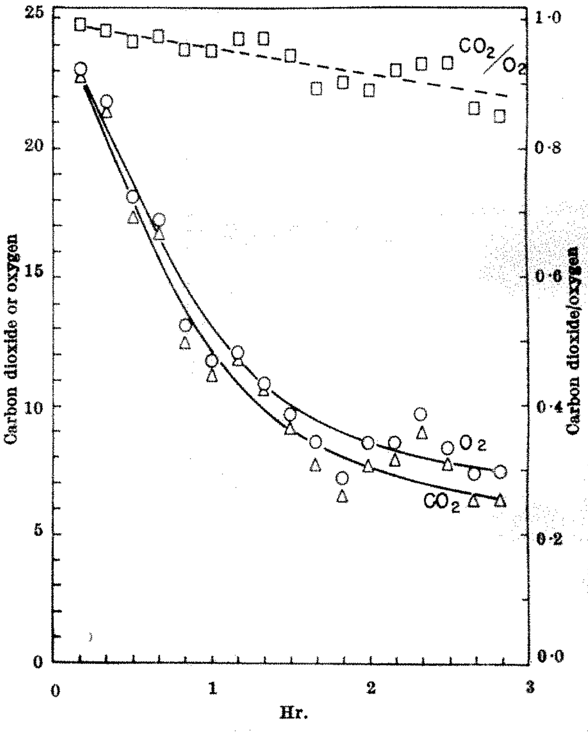

Fig. 2. Time course of the gas exchange during a photosynthetic experiment in a steady-state suspension of *Chlorella* 7–11–05. Carbon dioxide consumption and oxygen evolution in mm.^{3} of gas per mm.^{3} of cells per 10-min. intervals. Assimilatory quotients (carbon dioxide/oxygen) for the corresponding intervals. Note absence of recovery of rates

in such a suspension of older cells in later developmental stages results in a curve described in Fig. 2.

A basic observation from these studies is that the decrease in the rate of the photosynthetic gas exchange occurs at illuminance assumed to be most favourable for photosynthesis—just above light-saturating intensity. This decrease has nothing to do with the "lack of essential nutrient salts"[1] and it is not "the result of injury by the phosphate solution"[1] since the inhibition can be followed by a recovery of the rate without any apparent change in experimental conditions. The recovery of the rates is not connected with growth phenomena in the common sense of the word. Under conditions employed in these investigations, both these processes—the inhibition of photosynthesis and the increase in the rate as the result of building up some participants of the photosynthetic process—go simultaneously, the actual rate of the gas exchange being a consequence of the inter-play of these participating processes.

The magnitude of these two processes, and, therefore, the outcome of the competition between them, is greatly affected by the time factor in the course of the experiment and by the developmental stage of the cells. A resolution of the intricate intimacy of photosynthesis and development may prove advantageous in studies of both these processes.

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[1] Emerson, R., Cold Spring Harbor Symp. on Quant. Biol., **3**, 128 (1935).

[2] Myers, J., and Burr, G., *J. Gen. Physiol.*, **24**, 45 (1940).

[3] Steeman Nielsen, E., *Physiol. Plantarum*, **2**, 247 (1949).

[4] Kok, B., *Biochim. Biophys. Acta*, **21**, 234 (1956).

[5] Sironval, C., and Kandler, O., *Biochim. Biophys. Acta*, **29**, 359 (1958).

[6] Tamiya, H., Iwamura, T., Shibata, K., Hase, E., and Nihei, T., *Biochim. Biophys. Acta*, **12**, 23 (1953).

[7] Sorokin, C., *Physiol. Plantarum*, **10**, 659 (1957).

[8] Sorokin, C., and Myers, J., *J. Gen. Physiol.*, **40**, 579 (1957).

[9] Sorokin, C., and Krauss, R., *Proc. U.S. Nat. Acad. Sci.*, **45**, 1740 (1959).

[10] Iwamura, T., Hase, E., Morimura, Y., and Tamiya, H., "A. I. Virtanen Homage Volume. Biochemistry of Nitrogen" (Helsinki, 1955).

[11] Hase, E., Morimura, Y., and Tamiya, H., *Arch. Biochem. Biophys.*, **69**, 149 (1957).

[12] Pirson, A., Lorenzen, H., and Koepper, A., *Plant Physiol.*, **34**, 353 (1959).

[13] Sorokin, C., *Physiol. Plantarum*, **11**, 275 (1958).

[14] Sorokin, C., *Nature*, **184**, 613 (1959).

PATHOLOGY

Effect of Salicylate on the Activity of Plasma Enzymes in the Rabbit

ELEVATIONS of glutamic-oxaloacetic and glutamic-pyruvic transaminases have been reported to occur in sera from more than 50 per cent of children with rheumatic fever treated with either aspirin or sodium salicylate[1]. The injection of sodium salicylate in the rat caused an increased activity of xanthine dehydrogenase in the blood but a decreased activity of the enzyme in the liver, and it was suggested that the salicylate caused liver and other tissue damage leading to an increased rate of release of enzyme protein into the circulation[2]. We have therefore studied the effects of prolonged oral administration of salicylate on the activities of five plasma enzymes and on tissue histology in the rabbit.

Eight adult rabbits were given salicylate, in three divided doses each day, as either 5-grain soluble aspirin tablets B.P. or as 1 gm. per ml. aqueous solution of sodium salicylate. The aspirin tablets or the sodium salicylate solution were either placed or delivered from a syringe at the back of the animal's tongue and were readily swallowed when the mouth was then held shut. After an initial control period of two weeks, each rabbit received 0·75, 1·0 or 1·5 mgm. of the aspirin or sodium salicylate per gm. of body-weight each day for successive two-week periods. Blood specimens were collected from ear veins into heparinized containers at twice-weekly intervals and the separated plasma analysed for salicylates[3], glutamic-oxaloacetic and glutamic-pyruvic transaminases[4], *iso*citric dehydrogenase[5], lactic dehydrogenase[6] and aldolase[7]. The rabbits were killed by intravenous nembutal and portions of the liver and other tissues were removed and fixed in formol-saline for histological examination. Normal values for the activities of the five plasma enzymes in the rabbit were established by analysing random blood samples from 25 adult animals (Table 1).

Table 1. PLASMA ENZYME ACTIVITIES IN 25 CONTROL RABBITS
Results given as means and standard deviations

Enzyme	Units per ml. of plasma	Incubation temperature (° C.)
Glutamic oxaloacetic transaminase (*GOT*)	15·3 ± 4·6	37
Glutamic-pyruvic transaminase (*GPT*)	22·0 ± 8·5	37
Lactic dehydrogenase (*LD*)	5·2 ± 1·9	25
*iso*Citric dehydrogenase (*ICD*)	270 ± 90	25
Aldolase (*Ald.*)	10·8 ± 4·9	37

Increases in the activity of plasma enzymes were observed in six rabbits during the experiment, and typical results from two animals showing a marked and progressive rise in all the enzyme activities are given in Table 2.

Table 2. ACTIVITIES OF PLASMA ENZYMES IN TWO RABBITS

Day of experiment	Plasma salicylate (mgm./ 100 ml.)	Activity of plasma enzymes (units/ml.) *GOT*	*GPT*	*LD*	*ICD*	*Ald.*
30	52	9	12	15	280	8
32	64	11	8	34	660	6
37	78	58	18	60	820	73
40	75	203	25	132	3,200	150
19	38	5	3	4	160	9
24	52	7	4	12	170	5
27	31	26	40	12	2,600	34
29	52	780	140	32	4,000	125

It was necessary both to achieve and maintain plasma salicylate concentrations higher than 30 mgm. per 100 ml. in the susceptible rabbits to produce increased activity of plasma enzymes, and the time intervals between the initiation of salicylate administration and the first appearance of an enzyme abnormality ranged from 13 to 33 days. Histological examination revealed unequivocal signs of liver damage (cloudy swelling and fatty infiltration) in two of the six rabbits showing increased activity of plasma enzymes. The other tissues of these two rabbits and the tissues of the remaining animals were histologically normal.

The results show that the prolonged oral administration of salicylate caused increased activity of plasma enzymes in 6 out of 8 rabbits, but this was associated with definite signs of liver damage in only 2 of the animals. It is possible that these two rabbits represent an extreme case and that the liver and other tissue damage in the remaining animals was not gross enough to be detected by conventional histological methods. Zierler[8] has observed an increased flow of aldolase across rat muscle under conditions, such as anoxia, which alter muscle metabolism and membrane permeability. Concentrations of salicylate greater than 30 mgm. per 100 ml. uncouple oxidative phosphorylation reactions in mitochondrial suspensions and in the isolated rat diaphragm[9] and thus inhibit the synthesis of adenosine triphosphate. The selective permeability of cellular membranes depends on a normal rate of production of adenosine triphosphate. The uncoupling action of salicylate may therefore affect the permeability of many tissues, leading to an increased flow of enzyme proteins into the blood without producing structural damage to the tissues of such a degree that it would necessarily be seen during the histological examination of fixed tissues.

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[1] Manso, C., Taranta, A., and Nydick, I., *Proc. Soc. Exp. Biol. N.Y.*, **93**, 84 (1956).
[2] Mitidieri, E., and Affonso, O. R., *Nature*, **183**, 471 (1959).
[3] Trinder, P., *Biochem. J.*, **57**, 301 (1954).
[4] Reitman, S., and Frankel, S., *Amer. J. Clin. Path.*, **28**, 56 (1957).
[5] Wolfson, S. K., and Williams-Ashman, H. G., *Proc. Soc. Exp. Biol. N.Y.*, **96**, 321 (1957).
[6] Kaltenback, J. P., Becker, J. F., and Bernstein, I., *Amer. J. Clin. Path.*, **27**, 309 (1957).
[7] Sibley, J. A., and Lehninger, A. L., *J. Biol. Chem.*, **177**, 859 (1949).
[8] Zierler, K. L., *Ann. N.Y. Acad. Sci.*, **75**, 227 (1958).
[9] Brody, T. M., *Pharmacol. Rev.*, **7**, 335 (1955). Smith, M. J. H., and Jeffrey, S. W., *Biochem. J.*, **64**, 589 (1956).

Intracellular Potassium in Dietary Liver Necrosis

THIN slices of liver from rats fed a normal diet (M.R.C. 41*B* cubes) lose potassium when placed in cold saline solution and re-accumulate potassium against a concentration gradient when placed in warm oxygenated Ringer solution. Mudge[1] has shown a similar sequence of events to take place in rabbit kidney slices, and Aebi[2] has shown that rat liver slices can maintain a concentration gradient between internal potassium and the incubation medium if the incubation conditions are favourable.

Dietary liver necrosis is a condition which overtakes young rats fed the diet of Himsworth[3]. The diet consists mainly of yeast and starch and is severely deficient in α-tocopherol. Liver necrosis occurs suddenly about 17 days after weaning the rats to the yeast diet[4]. Apart from mild fatty infiltration the livers are histologically normal until necrosis occurs. Feeding α-tocopherol prevents necrosis. In the present investigation liver slices from rats fed the necrogenic diet of Himsworth were found to be unable to re-accumulate potassium when placed in warm oxygenated Ringer solution after losing potassium in a cold saline solution.

Table 1. POTASSIUM CONTENT OF LIVER SLICES AFTER INCUBATION IN THE COLD FOR 35 MIN. AND THEN IN WARM OXYGENATED RINGER FOR 60–80 MIN.

Diet	No. of animals	Potassium content (m.equiv./kgm. dry weight)
Stock or yeast + α-tocopherol	5	300 ± 30
Yeast diet 3–6 days	4	116 ± 13
Yeast diet 7 or more days	8	70 ± 18

Histologically normal livers from animals fed the necrogenic diet are referred to as prenecrotic livers, and the livers of animals which had been fed the diet for more than 9 days are considered as being in the late prenecrotic stage.

Pieces of liver and liver slices were dried to find water content, and then extracted with 0·15 *N* nitric acid. Sodium and potassium were measured in this extract with a flame photometer[5].

The livers of rats fed the necrogenic diet have a potassium content similar to that of livers of rats fed stock diet, or fed the necrogenic diet plus α-tocopherol. In frankly necrotic livers the potassium content is low.

Slices from prenecrotic livers and normal livers lose similar amounts of potassium when placed in a cold saline solution (sodium chloride, 150 m.equiv./l., potassium chloride, 5 m.equiv./l. at 0·5° C.). But when slices from prenecrotic livers are then placed in warm oxygenated Ringer solution they differ from slices of normal livers in showing a progressive failure to re-accumulate potassium (Fig. 1, Table 1). (Ringer phosphate, ref. 6, with 1·2 m*M* calcium and 6·6 m*M* potassium made up with water first distilled and then deionized in a 'Permutit' mixed-bed ion-exchange resin column is used as incubation medium.)

This inability is noticeable in slices from rats fed the diet for three days and becomes more severe as the time of feeding the diet increases.

The difference between slices from prenecrotic and from control livers can be detected in the first 20 min. of incubation. Slices from late prenecrotic livers

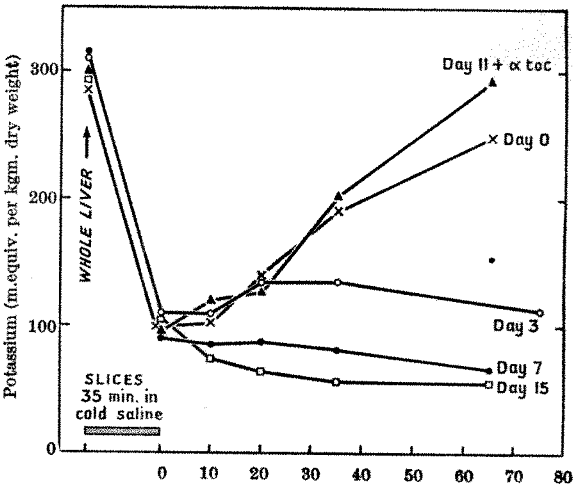

Fig. 1. Potassium accumulation in liver slices. Slices from rats fed necrogenic diet for increasing number of days. Day 11 + α-toc.: rat given 3 mgm. α-tocopherol 24 hr. before being killed. All slices kept in saline at 0° C. for 35 min. and then incubated in warm oxygenated Ringer solution

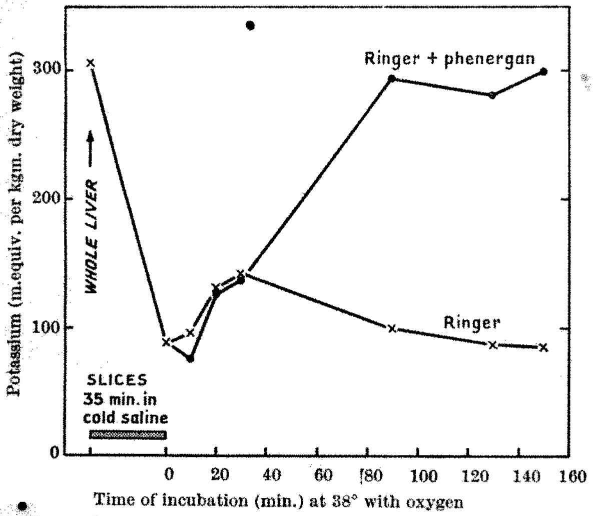

Fig. 2. Potassium accumulation in liver slices. Rat fed necrogenic diet for 5 days. Slices kept in saline at 0° C. for 35 min. and then incubated in warm oxygenated Ringer solution, with and without addition of 10^{-4} *M* 'Phenergan'

show a decline in oxygen uptake after the first 30 min. of incubation[4,7].

If a rat in the late prenecrotic stage is given α-tocopherol, liver slices taken 24 hr. later can accumulate potassium, like liver slices from rats fed stock diet, and oxygen uptake of slices does not decline. If slices from prenecrotic livers are incubated in Ringer solution containing 10^{-4} *M* 'Phenergan' (10-(2-dimethylamino*iso*propyl)phenothiazine hydrochloride), potassium accumulation is restored nearly to normal and oxygen uptake does not decline[4] (Fig. 2).

Sodium and water content of all these slices alter together and in a reciprocal manner to potassium content.

Homogenates of prenecrotic livers carry out oxidative phosphorylation with normal phosphorus/oxygen ratios.

Since the failure to re-accumulate potassium occurs in the early days of feeding the necrogenic diet, and since this failure occurs in the first 30 min. of incubation, the failure of oxygen uptake which occurs later may well be secondary to changes of electrolyte and water in the slices.

It is suggested, therefore, that these slices from prenecrotic livers probably exhibit the basic cell defect of dietary liver necrosis, namely, an inability to re-accumulate potassium once the internal cell structure has been disturbed by trauma, such as cold. The physiological source of trauma which causes *in vivo* necrosis is being sought.

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[1] Mudge, G. H., *Amer. J. Physiol.*, **167**, 206 (1951).

[2] Aebi, H., *Helv. Physiol. Pharmacol. Acta*, **11**, 96 (1953).

[3] Lindan, O., and Himsworth, H. P., *Brit. J. Exp. Path.*, **31**, 651 (1950).

[4] McLean, A. E. M., *Nature*, **185**, 191 (1960).

[5] Heckmann, K. D., and Parsons, D. S., *Biochim. Biophys. Acta*, **36**, 203 (1959).

[6] Cohen, P. P., in "Manometric Techniques and Tissue Metabolism", 119, edit. Umbreit (Burgess, Minneapolis, 1949).

[7] Chernick, S. S., Moe, J. G., Rodnan, G. P., and Schwarz, K., *J. Biol. Chem.*, **217**, 829 (1955).

Pathogenesis of Thrombosis

THE pathogenesis of thrombosis in man is still to a considerable extent a mystery despite the considerable advances in knowledge of the subject in recent years.

In 1924 Aschoff postulated the following mechanisms as being important in intravascular clotting: (1) Changes in the coagulability of the blood, (2) changes in the formed elements, (3) changes in the blood flow, (4) changes in the blood vessels.

To these may be added several other factors more or less represented in Aschoff's scheme: the ingestion of fats[1] or alternatively essential fatty acid deficiency[2]; ageing; stress; lack of exercise; female sex hormone; tobacco smoking; operations and pregnancy. The exact importance of several of these factors is still in a good deal of doubt, however.

Changes in the coagulability of the blood have been measured by clotting time tests by several methods, by many authors, in an endeavour to determine the presence of accelerated clotting in thrombosis, with variable results. Significant shortening of the heparin plasma clotting time within 48 hr. of the onset of vascular occlusion was reported by Poller[3]. The precise relationship of such accelerated clotting to vascular thrombus formation is not established and it has been suggested that it may be, in part at least, secondary, due to the release into the circulation of coagulation products activated during clotting[4].

Studies of more specific aspects of the coagulation pattern have been performed by numerous investigators. Increases of prothrombin activity[5], factor VII [6], fibrinogen[7], antithrombin[8], thromboplastin generation[6,7] and reduced fibrinolytic activity[9] have been reported in recent thrombosis.

Changes in the formed elements have included the measurement of platelet 'stickiness', which has been found to be increased by several authors.

Changes in the blood flow have not been extensively studied owing to the difficulty of simulating events *in vivo*.

The principal anatomical vascular change predisposing to thrombosis is atherosclerosis. Duguid[10] believes organization of intra-arterial clots and fibrin deposits to be a major cause of this.

The following simple experiment may be related to the third and fourth mechanisms postulated by Aschoff[11]: 20 ml. venous blood was collected into a siliconed syringe with the minimum of hæmostasis. It was then transferred to two wax-stoppered siliconed tubes, 10 ml. being placed in each. Instead of being allowed to clot in the normal way in the rack of a water-bath the tubes of blood were transferred to an electrical blood-mixer which rotates slowly at 30 revolutions per min., imparting a slow to-and-fro motion to the blood which may simulate the pulsatile blood flow in an elastic artery like the aorta.

Under these conditions, instead of a solid clot forming, coagulation took the form of a fine deposit of fibrin on the walls of the tubes. One of the tubes was removed from the mixer after 2 hr. and transferred to a rack in a water-bath. In this a visible solid clot formed. The other tube was left in the mixer for 12 hr., but in this instance a solid clot never formed on removal to a water-bath.

If this experiment simulates at all the events *in vivo* the following conclusions may be drawn: (1) blood clotting in a major vessel with good circulatory blood flow takes the form of fibrin deposition on the walls, which supports Duguid's 'thrombogenic' theory of atherosclerosis; (2) that a firm solid clot only forms in an artery when the local pulsatile blood flow is arrested.

The experiment is probably not related to events in venous thrombosis as the blood flow is sluggish, non-pulsatile, and atherosclerosis absent.

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[1] Fullerton, H. W., Davie, W. J. A., and Anastasopoulos, A., *Brit. Med. J.*, ii, 250 (1953).
[2] Sinclair, H. M., *Lancet*, i, 381 (1956).
[3] Poller, L., *Clin. Sci.*, **15**, 55 (1956).
[4] Poller, L., *J. Clin. Path.*, **12**, 331 (1959).
[5] Sandrock, R. S., and Mahoney, E. B., *Ann. Surg.*, **128**, 521 (1948).
[6] Poller, L., *J. Clin. Path.*, **10**, 348 (1957).
[7] McDonald, L., European Congress of Hæmatology, London (abstract) (1959).
[8] Oschner, A., Kay, J. H., deCamp, P. J., Hutton, S. P., and Balla, G. A., *Ann. Surg.*, **131**, 652 (1950).
[9] Hume, R., *Brit. Heart J.*, **20**, 15 (1958).
[10] Duguid, J. B., *J. Path. Bact.*, **58**, 207 (1946).
[11] Aschoff, L., "Lectures in Pathology" (New York, 1924).

ANATOMY

Marchi-Positive Bodies in Human Globus Pallidus

THE Marchi method has been used extensively in this laboratory in the course of studies on degenerating nerve fibres in the central nervous system made during the past twelve years. The purpose of this communication is to report the invariable occurrence of black, Marchi-positive bodies in the globus pallidus in brains examined by this method. This is an incidental finding, unconnected with the nerve fibre degeneration under study. No previous report of it has been found in the literature.

More than 250 brains have been examined by this method. These brains came from patients dying from a wide variety of neurological and non-neurological conditions. In every brain examined there is black staining in the pallida, the amount of staining varying from brain to brain, but being approximately the same on the two sides in each brain. This black staining is in the form of rounded 'bodies' of varying size arranged in groups, frequently consisting of one or two larger 'bodies' surrounded by a number of small 'bodies'; their diameter varies from 1μ to 60μ (Figs. 1 and 2). The 'bodies' are not intracellular and are not enclosed within the sheaths of nerve fibres. Their appearance is the same in horizontal, oblique and coronal planes of section.

Sections from blocks next to those prepared by the Marchi method have been stained by the usual variety of neuro-histological methods to show changes in neurones, nerve fibres and glia. A few pink 'droplets' have occasionally been seen in sharlach *R* preparations which might be the equivalent of some of the Marchi-stained 'bodies', but otherwise no such 'bodies' have been seen in sections stained by any other method.

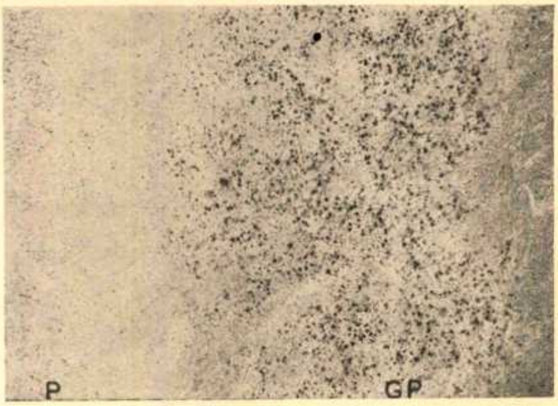

Fig. 1. Marchi preparation of putamen (*P*) and globus pallidus (*GP*) from normal brain, showing black, Marchi-positive 'bodies' in globus pallidus (× *c*. 13·5)

The purpose of the Marchi method is to demonstrate the presence of degenerating myelin. The essential mechanism in the method is the reduction of osmium tetroxide to osmium dioxide, which is black. Both normal and degenerating myelin cause this reduction; but it is inhibited in normal myelin by the addition of potassium dichromate or chlorate to the osmium tetroxide. Thus, in principle, only degenerating myelin stains black. In practice, artefacts of various kind may occur with this method. It must be stressed that the 'bodies' now described are completely unlike any of the artefacts ever seen, and that they are completely unlike any form of black staining in the central nervous system known to be associated with the degeneration of myelinated nerve fibres.

These bodies have a constancy of 'structure' which makes recognition of the globus pallidus easy even in tiny pieces of basal ganglia. No such 'bodies' have been found in the putamen, but very occasionally a very small number are present in the adjoining part of the internal capsule and/or in the more cranial part of the substantia nigra.

Various factors have been considered with reference to the occurrence and number of the 'bodies'. More than 250 brains have been examined by the Marchi method. Neoplastic disease, most frequently of the pelvic organs, was the most common cause of death, but brains from patients who died because of cardiac disease, of various forms of accident with and without head injury, pneumonia, tuberculosis, poliomyelitis, polyneuritis and different degenerative diseases of the nervous system were all examined. As mentioned already, the 'bodies' in the globus pallidus varied in number from brain to brain, but there were some present in all brains. No correlation could be established between the cause of death and the abundance

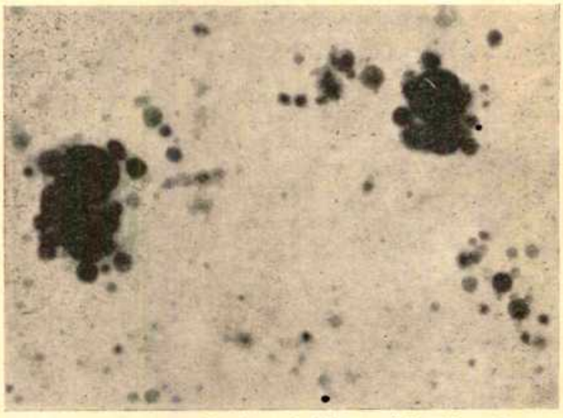

Fig. 2. Higher magnification of globus pallidus from Fig. 1, showing grouping of black 'bodies' (× *c*. 600)

of the 'bodies'. Similarly, no correlation could be established between the age of the patients, which varied from 20 to more than 70 years, and the number of the 'bodies'.

Although unlikely, the question might be raised whether the post-mortem treatment of the brains might have something to do with the occurrence and number of the 'bodies'. The interval between death and post-mortem varied from 2 hr. to 3 days. As the brains came not only from this hospital but from many other hospitals throughout the country, the possible effect of variations in the techniques of examination and of fixation can be excluded. No correlation could be established between the interval between death and fixation and the number of the 'bodies'.

The brains from a few species other than man have been examined by the Marchi method. These came from rhesus monkeys, cats, horses, sheep, guinea pigs, rats and mice. In none of these brains were any of these 'bodies' seen. However, the number of animals examined was too small for any definite conclusion to be drawn about the absence of these 'bodies' in mammals other than man.

At present no conclusion as to the nature of these 'bodies' is possible; but it is tentatively suggested that they may indicate some biochemical feature characteristic of the globus pallidus which has not yet been identified. Much interest is now focused on the basal ganglia. It is possible that with increasing knowledge of this region the significance of the findings reported here will become recognized.

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BIOLOGY

Dorsal Fontanelles in Selachian Neurocrania

Since the classical work of Gegenbaur[1] and Parker[2] the anterior opening lying on the dorsal surface of the selachian neurocrania is generally termed 'anterior fontanelle' in spite of the fact that it is different in its morphological significance and relations in different selachians. In some forms, posterior to this fontanelle, a small opening, the epiphysial foramen, is found; and in others another fontanelle, which is generally termed the 'supracranial fontanelle', appears. The study of several species of several families of Selachii showed that it is of great importance to name such fontanelles according to their morphological relation with the cranial cavity.

In *Squalus blainvillii* the fontanelle lies beyond the morphological anterior wall of the cranial cavity and represents the dorsal opening of the cavum præcerebrale found inside the spout-shaped rostral cartilage, and is accordingly named the precerebral fontanelle. Posterior to this fontanelle a small epiphysial foramen occurs. In *Squatina oculata* the fontanelle is completely supracranial, as the morphological anterior wall of the cranial cavity extends to its anterior margin. Behind it is a small epiphysial foramen. In *Triaenodon obesus* the brain extends anteriorly beyond the posterior edge of the fontanelle and thus it can be differentiated into two morphologically different parts: the anterior supracranial fontanelle lying above the cranial cavity, and the precerebral fontanelle representing the dorsal opening of the cavum præcerebrale enclosed between the three rostral cartilages. In *Torpedo sinus* the fontanelle is morphologically similar to that of *Triaenodon*, and posterior to it lies the epiphysial foramen. In *Raja batis* there are two large fontanelles separated by a narrow cartilaginous bridge. The anterior is differentiated into a precerebral fontanelle leading to the cavum præcerebrale inside the rostral cartilage, and an anterior supracranial fontanelle. The posterior is completely supracranial and is, here, termed the posterior supracranial fontanelle to differentiate it from the anterior one. In *Taeniura forskali* is found only one large fontanelle, which is completely supracranial, and termed the supracranial fontanelle.

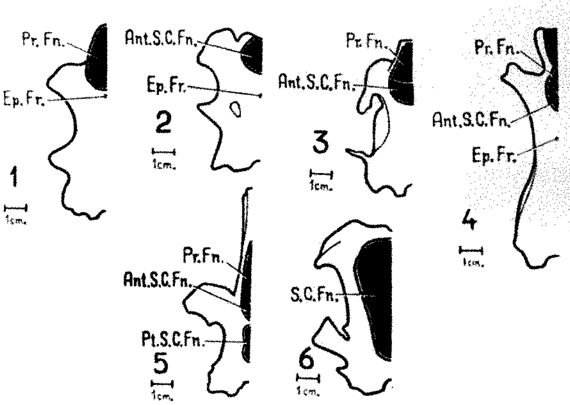

Fig. 1. Dorsal view of the neurocranium of: 1, *Squalus blainvillii*; 2, *Squatina oculata*; 3, *Triaenodon obesus*; 4, *Torpedo sinus*; 5, *Raja batis*; 6, *Taeniura forskali*. *Ant.S.C.Fn.*, Anterior supracranial fontanelle; *Ep.Fr.*, epiphysial foramen; *Pr.Fn.*, precerebral fontanelle; *Pt.S.C.Fn.*, posterior supracranial fontanelle; *S.C.Fn.*, supracranial fontanelle

Such comparative study of the dorsal fontanelles in different selachian neurocrania shows that when rostral cartilages are not present the fontanelles are morphologically supracranial as in the case of *Squatina* and *Taeniura*. In cases where rostral cartilages are present a precerebral fontanelle is represented, and it may be the only fontanelle present as in *Squalus*, or it may be confluent with a supracranial fontanelle as in *Triaenodon*, *Torpedo* and *Raja*. This view can be substantiated by the embryological work of El-Toubi[3] and Hamdy[4], who show that the single, spout-shaped rostral cartilage develops from three strips of cartilage having the same relations as the triradiate rostral cartilages. Thus it is easy to homologize the whole fontanelle in *Squalus* with only the precerebral fontanelle in *Triaenodon*, *Torpedo* and *Raja*; the anterior supracranial fontanelle in *Triaenodon*, *Torpedo* and *Raja* with that of *Squatina*; and the whole fontanelle in *Taeniura* with both the anterior and posterior supracranial fontanelles in *Raja*, being confluent with each other in consequence of the absence of the cartilaginous bridge separating them in the latter fish.

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[1] Gegenbaur, C., "Das Kopfskelet der Selachier" (Leipzig, 1872).
[2] Parker, W. K., *Trans. Zool. Soc.*, **10**, 189 (1879).
[3] El-Toubi, M. R., *J. Morph.*, **84**, 227 (1949).
[4] Hamdy, A. R., *Proc. Egypt. Acad. Sci.*, **13** (1957/58).

Spore Release in *Peronospora tabacina* Adam.

DOWNY mildew or blue mould of tobacco caused by *Peronospora tabacina* Adam. is destructive in the American and Australian regions and has now been reported for the first time in Europe[1]. It survives in association with over-wintering or over-summering host plants of which infected *N. tabacum* is of major importance[2,3]. Such plants produce conidia that are airborne and can establish infection in distant areas[2].

De Bary suggested that dissemination of conidia of *Phytophthora infestans* (Mont.) de B. was brought about by changes in humidity causing violent movement of conidiophores and conidia were "thrown off in every direction"[4]. Ingold[5] did not agree with de Bary's conclusion ; but Pinckard[6], working with *Peronospora tabacina*, supported it and also reported "ejection of sporangia (conidia) from their sterigmata by energy applied at the point of sporangial attachment". Hirst[7] concluded that sporangia (of *P. infestans*) were not removed by wet or dry gales but were dislodged by wetting, rapid changes in relative humidity and strong light. All investigators associated conidiophore movement with an increasing or decreasing relative humidity and spore dissemination with decreasing humidity. Cruickshank[8] reported intensity of spore discharge in *P. tabacina* with change in relative humidity and mechanical shock. In the experiments reported here, factors other than ambient relative humidity were also concerned in spore release.

Dissemination of conidia was investigated at Canberra, Australia, with the aid of a spore trap[9] operated among tobacco plants showing heavy sporulation. In three seasons of field experiments, the number of conidia trapped each hour of the day increased to a maximum between 0930 and 1100 hr. for periods of 12–16 consecutive days. This daily cycle was not interrupted by overnight relative humidity too low for sporulation, the conidia disseminated having been produced at an earlier date. Thus spore release was not conditional on a decreasing humidity being preceded by overnight saturation. This conclusion was confirmed by glasshouse tests in which the relative humidity subsequent to sporulation was maintained at low levels for long periods (Table 1 and Fig. 1). The results of numerous tests show that the number of days in the daily cycle of spore release was least when there was a wide range, and repeated sudden fluctuations, in temperature and humidity. Daily periodicity for 4–10 days was common.

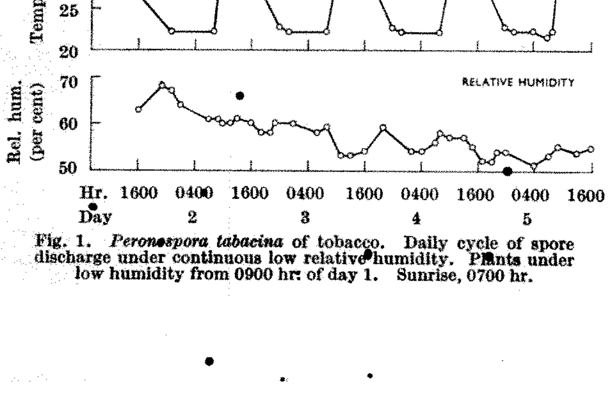

Fig. 1. *Peronospora tabacina* of tobacco. Daily cycle of spore discharge under continuous low relative humidity. Plants under low humidity from 0900 hr. of day 1. Sunrise, 0700 hr.

Table 1. RELATIVE HUMIDITY DURING CONSECUTIVE DAYS AND AT TIMES WHEN NUMBERS OF CONIDIA TRAPPED WERE INCREASING

Time of rise in numbers of conidia trapped (hr.)	Relative humidity (per cent)*			Period of test (consecutive days)
	Means at hours in first column	Actual maximum and minimum for total period of days		
		Max.	Min.	
0712–1300	61·8–63·4	74	55	10†
0712–1112	57·0–48·2	79	42	5
0815–1045	47·5–42·5	59	37	4
0745–1120	51·2–51·0	60	37	4
0645–0930	57·5–57·2	68	51	4‡

* From hygrographs among sporulating plants.
† Shaded glasshouse.
‡ Also shown graphically.

Examination of the fruiting structures of the pathogen *in situ* showed that twisting movements of the conidiophores were due to loss of turgidity in the stalk. This was followed by collapse of the branched superstructure, with many conidia still attached to it, on to the leaf surface. Spore release ceased but was resumed from both living and dead leaves on the same or succeeding days following periods of high ambient humidity. The spore trap data also showed that rising temperature, or light following a period of darkness, were associated with spore release. Collapsed conidiophores with attached conidia remained in that state, subsequent movements being restricted to some of the branches. The only method of spore dispersal that occurred either with or without conidiophore movement was, as described by Pinckard, due to "energy applied at the point of attachment".

These results show that spore release is regulated to provide for dissemination at intervals in time. Factors that appear to be responsible for spore release—reduction in relative humidity, increase in temperature, or light—could cause humidity or temperature-humidity changes in the region occupied by the conidiophores and thus ensure a progressive release of conidia. Whatever the mechanism involved, the results clearly show that spore discharge is a response to stimuli repeated at intervals, and that the normal interval was approximately 24 hr. Under this arrangement, conidia were held ready for dispersal and could take advantage of favourable conditions occurring at any time over a period of days following their production.

A full account of the work will be presented elsewhere.

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[1] "Seedborne Diseases of Tropical and Subtropical Crops", 8 (European and Mediterranean Plant Protection Organ., Jan. 1959).
[2] McGrath, H., and Miller, P. R., *Plant Dis. Rep.*, Supp. 250 (1958).
[3] Hill, A. V., C.S.I.R.O. (Aust.) Div. of Plant Industry, Tech. Paper 9 (1957).
[4] Waggoner, P. E., and Taylor, G. S., *Phytopath.*, **48**, 46 (1958).
[5] Ingold, C. T., "Spore Discharge in Land Plants" (Oxford, 1939).
[6] Pinckard, J. A., *Phytopath.*, **32**, 505 (1942).
[7] Hirst, J. M., *Outlook on Agric.*, **2**, 16 (1958).
[8] Cruickshank, I. A. M., *Aust. J. Biol. Sci.*, **11**, 162 (1958).
[9] Hirst, J. M., *Ann. App. Biol.*, **39**, 257 (1952).

The Lungworm Protostrongylus rufescens in Australia

Of the lungworms described from domestic sheep only two, namely, *Dictyocaulus filaria* and *Muellerius capillaris*, have been identified in Australia[1], despite careful searching in the eastern States[2]. On one occasion a single larva of *Protostrongylus* spp. was found in fæces of a sheep in Tasmania (Gordon, H. McL., personal communication), but earlier records of the adults are regarded as mistaken[1,2].

P. rufescens has now definitely been identified in lungs of sheep from Millicent, in the south-eastern part of South Australia, and its larvæ in fæces from the flock. Larvæ have also been found in the abomasal contents of a sheep from Kojonup in the south-west of Western Australia. The indications from the large numbers of adults recovered (up to 600 in a single sheep) are that the worm is by no means rare in the south-western part of Australia.

The anatomy conforms in detail with that described by Gerichter[3] except that the local adults, eggs and first-stage larvæ are rather smaller; for example, males are 18–30 mm. and females 28–45 mm. long, against Gerichter's 22–46 mm. and 30–65 mm. respectively. Other authorities[4] record dimensions comparable with our measurements.

The surface of the affected lungs showed scattered yellowish grey consolidated patches of irregular shape, 3–15 mm. in diameter, identical with those described by Kassai[5]. The worms were found mostly in clumps of up to ten, obstructing bronchioles, with some lying in larger air-vessels. A rapid and highly effective method of extracting the worms is to fill the trachea and bronchi with saline, tip up and down several times, pour out, and repeat the process a number of times.

The material from Kojonup was kindly made available by Dr. I. W. Parnell, of Perth. We wish to thank Mr. A. W. Banks for advice in connexion with this work.

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[1] Ross, I. C., and Gordon, H. McL., "The Internal Parasites and Parasitic Diseases of Sheep" (Angus and Robertson, Sydney, 1936).

[2] Seddon, H. R., "Diseases of Domestic Animals in Australia. Part 1. Helminth Infestations" (Government Printer, Sydney, 1950).

[3] Gerichter, C. B., *Parasitol.*, **41**, 166 (1951).

[4] Lapage, G., "Veterinary Parasitology" (Oliver and Boyd, London, 1956).

[5] Kassai, T., *Acta Vet. Lung*, **7**, 351 (1957).

MICROBIOLOGY

Feedback Control of Tryptophan Biosynthesis

Microbiologists are becoming increasingly interested in the regulation of biosynthesis. There has been a number of reports of normal metabolites inhibiting the action of enzymes necessary for their own biosynthesis[1–6], and others of the inhibition of enzyme formation or activity[7–9]. Umbarger[1,2] has called this type of inhibition 'negative feedback'.

In this laboratory we have been interested in tryptophan biosynthesis, particularly the identification of intermediates in the conversion of anthranilic acid to indoleglycerol phosphate[10]. Moyed and Friedman[6] have recently demonstrated that tryptophan can inhibit the metabolism of anthranilic acid by a cell-free preparation of *Escherichia coli*, and considered that the site of inhibition was the condensation of anthranilic acid with 5-phosphoribosyl-1-pyrophosphate. We considered that this interpretation was premature, since the preparation could carry out more than one step and because N-*o*-carboxyphenyl-D-ribosylamine, the unphosphorylated form of the postulated condensation product, is chemically unstable at biological *p*H (ref. 10 and unpublished work by Doy, C. H.). It seemed to us that the site of inhibition might equally well be the Amadori rearrangement step:

$$\begin{tikzcd} \text{Anthranilic acid} \arrow[r] & \text{N-}o\text{-carboxyphenyl-D-ribosylamine-5-phosphate} \arrow[l, dashed, bend left, "\text{decomposition}"] \arrow[d, "\text{inhibited}"] \\ & \text{1-(}o\text{-carboxyphenylamino)-1-deoxyribulose-5-phosphate} \end{tikzcd}$$

In any event we suspected that anthranilic acid was only formed by the appropriate tryptophan auxotrophs of *Aerobacter aerogenes* and *Escherichia coli* in the absence of growth.

We have now shown (Fig. 1) that *Aerobacter aerogenes* mutant strain *NCW-T-17A*, able to respond to indole, but not anthranilic, acid and known to accumulate anthranilic acid in washed-cell experiments with minimal substrates, does not excrete detectable amounts of anthranilic acid until growth on limiting L-tryptophan has ceased. The coincidence of end of growth and the onset of accumulation suggests that tryptophan is able to inhibit a process necessary for the biosynthesis of its own precursor anthranilic acid. Because there was no lag between end of growth and the excretion of anthranilic acid, the inhibition is likely to be on the action of an enzyme rather than on enzyme formation. This conclusion is supported since cells harvested during growth, washed to remove tryptophan and then transferred to minimal substrate, immediately begin to form anthranilic acid (Fig. 2, but note tendency for acceleration). In fact, they form the intermediate at a faster rate than that observed after growth (Fig. 1). This is probably because the fresh substrate represents a condition closer to the optimum.

The conclusion that tryptophan is able to control its own biosynthesis by inhibiting the action of an enzyme necessary for a stage prior to anthranilic acid suggests that the inhibition reported by Moyed and Friedman[6] is not the earliest site of action and is therefore more of a potential control point than a real one. Since nothing is known of specific tryptophan precursors prior to anthranilic acid, the inhibition now reported makes this part of the pathway particularly important. Because of this lack of knowledge it is also necessary to have reservations about the role of anthranilic acid, since the 'natural' condensation of 5-phosphoribosyl-1-pyrophosphate may be with a compound other than anthranilic acid[11].

Tryptophan auxotrophs grow well if supplied with L-tryptophan, which suggests that tryptophan does not inhibit the biosynthesis of precursors which it shares in common with phenylalanine and tyrosine. It is likely that, as with other examples[3,4], the 'end product' will be found to inhibit the earliest step specific to its own biosynthesis.

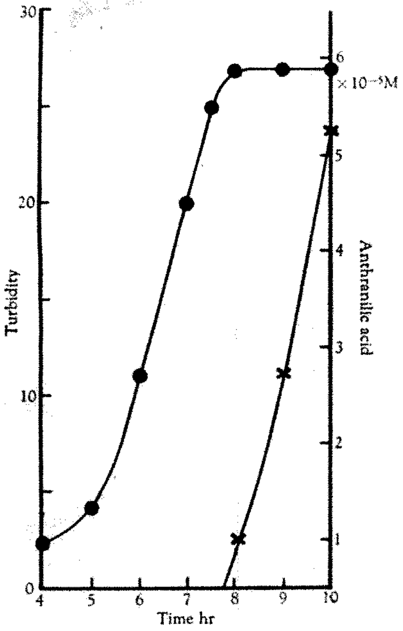

Fig. 1

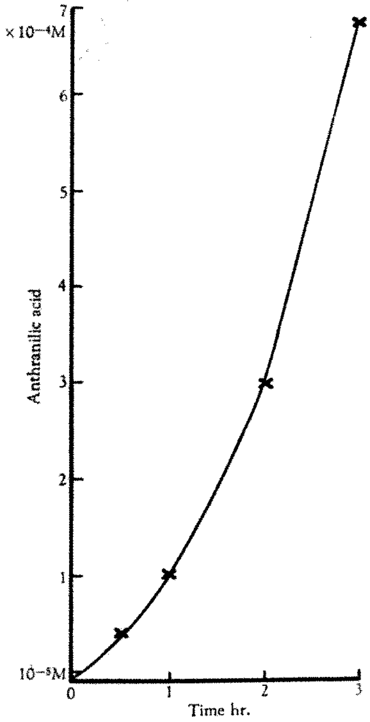

Fig. 2

Fig. 1. Growth of tryptophan auxotroph *Aerobacter aerogenes NCW-T-17A* compared with excretion of anthranilic acid. Minimal substrate, 15 ml. (disodium hydrogen phosphate, 12·8 gm.; potassium dihydrogen phosphate, 2·72 gm.; ammonium chloride, 5·4 gm.; D-glucose, 36 gm.; magnesium sulphate, 10^{-1} *M*, 2 ml.; made up to 2 l., *p*H 7·7) supplemented with 2×10^{-5} *M* L-tryptophan, was lightly inoculated with the test strain and shaken in a ⊥-tube at 37° C. Samples were taken for turbidity measurements (625 mμ filter, 20 = 0·6 mgm. dry wt./ml.) and determination of anthranilic acid by the method of Bratton and Marshall (ref. 12) (slightly less than 10^{-5} *M* was detectable). ●, Growth (turbidity); ×, anthranilic acid; none was detected until 8 hr.

Fig. 2. Growing cells (Fig. 1) harvested at turbidity 20, washed and resuspended in minimal substrate to equivalent turbidity, then shaken in ⊥-tube at 37° C.

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[1] Umbarger, H. E., *Science*, **123**, 848 (1956).
[2] Umbarger, H. E., and Brown, B., *J. Biol. Chem.*, **233**, 415 (1958).
[3] Strecker, H. J., *J. Biol. Chem.*, **225**, 825 (1957).
[4] Yates, R. A., and Pardee, A. B., *J. Biol. Chem.*, **221**, 757 (1956).
[5] Wyngaarden, J. B., and Ashton, D. M., *J. Biol. Chem.*, **234**, 1492 (1959).
[6] Moyed, H. S., and Friedman, M., *Science*, **129**, 968 (1959).
[7] Cohn, M., Cohen, G. N., and Monod, J., *C.R. Acad. Sci., Paris*, **236**, 746 (1953).
[8] Gorini, L., and Maas, W. K., *Biochim. Biophys. Acta*, **25**, 208 (1957).
[9] Wijesundera, S., and Woods, D. D., *Biochem. J.*, **55**, viii (1953).
[10] Doy, C. H., and Gibson, F., *Biochem. J.*, **72**, 586 (1959).
[11] Doy, C. H., "Tryptophan Biosynthesis in Micro-organisms" in "Rev. Pure App. Chem." (in the press).
[12] Bratton, A. C., and Marshall, E. K., *J. Biol. Chem.*, **128** 537 (1939).

Growth of Iron Bacteria in Crude Tar

In continuous distillation units of the Midland Tar Distillers, Ltd., in which vertical retort gas tar was distilled, it was found that certain heat exchangers pre-heating the crude tar became rapidly choked with deposits of fibrous material. It was at first thought that this was adventitious material such as asbestos or vegetable fibres present in the crude tar feed with entrained particles of solid material. The crude feed was, however, continuously filtered through a 'Rotoclene' filter with a clearance of ten-thousandths of an inch and examination of the material held back by this filter did not resemble the deposit within the heat exchanger. It was this fact, together with the quantity and consistent appearance of the deposit at one point in the heat exchanger train, that suggested, in fact, it was essentially a growth in the tar medium.

The fibrous matter in the deposit was demonstrated to be neither asbestos nor vegetable, and an analysis of the ashed deposit showed it to consist mainly of iron oxide and manganese salts. Under the microscope, iron oxide skeletons were readily discernible among the ash, which suggested that the deposit may at least in part have been due to biological growth. The ash was also strongly paramagnetic, which lent weight to this view, as the production of a magnetic form of iron oxide is characteristic of the activity of iron bacteria.

Samples of a freshly collected deposit were examined under the microscope. Dense populations of motile bacilli and cocci were observed, and, in addition, what appeared to be fungal filaments with multi-cellular structure with branching and spores at the branch ends were also noted. The diameter of the fibres was about 20 microns, which although larger than that of most observed filamentous bacteria, is still within the range of size of filamentous bacteria.

The suggestion, therefore, was that the prime reason for the formation of the deposit was the development of iron bacteria. Conditions in the heat exchanger were favourable for the growth of diverse species of micro-organisms. The temperature-range over the section in which the deposit was prevalent was 30–70° C.

When the quantity of iron present in the deposit was considered, it did not seem reasonable that this iron was a result of corrosion, because gross corrosion of the heat exchanger was not taking place. It seemed more reasonable to assume that the iron oxides had come from a concentration of the soluble iron salts present in the crude tar either by biological activity or purely physical and chemical means.

The deposit also contained 3·0 per cent (w/w) of sulphides, and as only a trace of sulphur was present in the crude tar feed (as thiosulphates, thiocyanates, sulphates), here again was a process of concentration to be explained either by biological or physical and chemical means.

The mild conditions in the heat exchangers and the significant temperature-range (30–70° C.) pointed to biological activity as the prime cause of the deposit.

The cultivation of iron bacteria outside their normal environment is difficult, but a reasonable amount of 'fibrous' organism was grown on a solid medium which included an aqueous extract of the deposited material. Although the filaments could not be positively identified, their appearance agreed with descriptions found in the literature of some of the iron bacteria. They did resemble the fibres actually present in the heat exchanger deposits, so far as magnetic properties, appearance and sulphide content were concerned.

It is possible that the small motile vibrio type of organisms observed under the microscope in fresh samples were sulphate-reducing bacteria, but this has not yet been confirmed.

I wish to thank Dr. N. J. Cartwright and Mr. F. W. Moore, of the Department of Bacteriology, University of Birmingham, for their interest and assistance on the microbiological side.

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Observations on the Nagler Reaction of some *Clostridia*

NAGLER[1] showed that the surface of colonies of *Clostridium oedematiens* type *A* growing on egg-yolk – sheep-blood agar, is covered by a pearly layer. Subsequent investigations by McClung *et al.*[2] and McClung and Toabe[3] showed that this phenomenon was not restricted to *Cl. oedematiens* type *A*, but was also shown by all types of *Cl. botulinum* and by *Cl. sporogenes*, and was more evident if the blood was omitted from the medium. All the *Clostridia* which show this pearly layer on egg-yolk agar also produce an opalescence within the medium, somewhat similar in appearance to that produced by anaerobes such as *Cl. welchii* and *Cl. bifermentans*, which produce lecithinase. Oakley *et al.*[4] showed that cultures and culture-filtrates of *Cl. oedematiens* type *A* produce two distinct types of opalescence in egg-yolk emulsions: a diffuse opalescence due to a hæmolytic lecithinase (*Cl. oedematiens* γ-toxin), and a more confined opalescence due to a lipase (*Cl. oedematiens* ε-antigen). They suggested that the pearly layer also produced by *Cl. oedematiens* type *A* might be caused by the lipolytic ε-antigen (see also ref. 5). Sussman (personal communication) showed that cultures of *Cl. sporogenes* were lipolytic when grown on tributyrin agar ('Oxoid'), and that this organism developed a pearly layer when grown on an agar medium containing triolein.

The present communication presents briefly the results obtained in a study of the pearly layers produced by *Cl. sporogenes*, all types of *Cl. botulinum* and *Cl. oedematiens* type *A*.

A number of clostridia were examined for lipolytic activity by growing them as surface cultures on tributyrin agar ('Oxoid') enriched with 5 per cent of Fields's extract of red cells. Lipolysis, indicated by the development of zones of clearing in the medium about areas of growth, was shown by strains of *Cl. sporogenes*, all types of *Cl. botulinum* and *Cl. oedematiens* type *A*, but not by strains of *Cl. bifermentans*, *Cl. welchii* (types *A–F*), *Cl. histolyticum* or *Cl. oedematiens* type *B* (see Table 1). Thus, of the species examined, only those which produce a pearly layer on egg-yolk media showed lipolytic activity on tributyrin agar.

Table 1. ACTION OF SOME *Clostridia* ON MEDIA WITH AND WITHOUT LECITHIN AND FAT

Organism	Clearing on 'Tributyrin' agar	Egg-yolk agar		Cream-milk agar		Fat-free milk agar	
		Opal.*	Pl.†	Opal.	Pl.	Opal.	Pl.
Cl. welchii (*A–F*)	–	+	–	–	–	–	–
Cl. bifermentans	–	+	–	–	–	–	–
Cl. histolyticum	–	–	–	–	–	–	–
Cl. botulinum (*A–E*)	+	+	+	+	+	–	–
Cl. sporogenes	+	+	+	+	+	–	–
Cl. oedematiens, type *A*	+	+	+	+	+	–	–
Cl. oedematiens, type *B*	–	+	–	–	–	–	–

* Opal., opalescence. † Pl., pearly layer.
+ or –, present or absent, respectively.

The various organisms were then cultured on egg-yolk agar, full-cream-milk agar and fat-free-milk agar. The results, which are summarized in Table 1, show that all the lipolytic organisms examined produced an opalescence and a pearly layer on fat-containing media, but not on the fat-free medium. Lecithinase activity was shown only on egg-yolk agar.

Egg-yolk agar plates, full-cream-milk agar plates and fat-free-milk agar plates were spotted with pancreatic lipase (Light and Co.) and incubated at 37° C. The egg-yolk and cream plates developed zones of opalescence covered by a pearly layer, similar in appearance to those produced by the lipolytic *Clostridia*. No opalescence or pearly layer was produced by pancreatic lipase on the fat-free medium. These plates, and all the plate cultures of the anaerobes, were flooded with a saturated solution of copper sulphate, allowed to stand for a few minutes, and then rinsed with water (the copper soap test for lipase). Positive results, shown by the appearance of bluish-green zones of precipitate in the medium, were given by the egg-yolk and cream plates treated with pancreatic lipase, and by all the cultures of *Clostridia* growing on egg-yolk and cream agar and producing a pearly layer. Negative results were given by cultures not associated with a pearly layer.

These results indicate that the pearly layer and opalescence produced on egg-yolk and other fat-containing media by the *Clostridia* examined are due to lipolytic activity, and probably consist of free fatty acid. No pearly layer is produced on tributyrin agar by lipolytic organisms because the products of lipolysis (glycerol and butyric acid) are soluble. Macfarlane suggested[6] that *Cl. oedematiens* type *D* (*Cl. hæmolyticum*) may produce traces of a lipase. This organism has not been studied in the present investigation.

If the conclusions drawn are correct, it seems likely that the butyrous scum which sometimes develops on the surface of cooked-meat broth cultures of *Cl. botulinum* and *Cl. sporogenes* consists of fatty acid liberated by lipolysis. Further, the absence of a lipase from cultures of *Cl. welchii* provides an explanation for the characteristic fatty appearance of the œdema fluid produced in pure *Cl. welchii* gas gangrene.

Further investigations along these lines are at present being made and will form the basis of a subsequent more detailed communication.

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[1] Nagler, F. P. O., *Nature*, **153**, 496 (1944); *Aust. J. Exp. Biol. Med. Sci.*, **23**, 59 (1945).

[2] McClung, L. S., Heidenreich, P., and Toabe, R., *J. Bact.*, **50**, 715 (1946).

[3] McClung, L. S., and Toabe, R., *J. Bact.*, **53**, 139 (1947).

[4] Oakley, C. L., Warrack, G. H., and Clarke, P. H., *J. Gen. Microbiol.*, **1**, 91 (1947).

[5] Macfarlane, M. G., *Biochem. J.*, **42**, 590 (1948).

[6] Macfarlane, M. G., *Biochem. J.*, **47**, 267 (1950).

Survival of Bacteria on Drying in Sugar/Protein Mixtures

In an investigation of the protection afforded by various sugars in the preservation of the viability of dried cultures, Hellier[1] directed attention to the advantage of a sucrose/peptone mixture over either sucrose or peptone alone. In 1951, Fry and Greaves[2] proposed their *mist. desiccans*, a serum/broth mixture containing 7·5 per cent glucose, and this has come into extensive use as a medium in which to dry a wide variety of organisms with a high retention of viability. In general, in the presence of either sugar or protein alone, the percentage survival is relatively poor; only in a mixture of the two in suitable proportions and in adequate concentration is a high degree of viability retained after drying and reconstitution. The underlying action of such mixtures has not hitherto been satisfactorily explained.

The concept of a bacterial cell as confined by a thin plasma membrane supported by an outer, stronger and more permeable cell wall is now well established, and the phenomenon of plasmolysis as displayed by various Gram-negative organisms on exposure to solutions of electrolytes and sugars such as glucose or sucrose has long been recognized as the result of the collapse of the plasma membrane due to osmotic forces caused by these substances, to which only the cell wall is permeable. During such plasmolysis the overall shape of the cell as defined by the cell wall is still clearly seen.

By the use of a micro-culture chamber[3] we have observed that cells of *Bacterium coli* dried in various sugar solutions, or even after exposure for some time to their concentrated solutions, exhibit, on returning to ordinary growth media such as broth or peptone water, a high proportion of spherical forms or protoplasts resulting from the rupture of the cell wall by the high diffusion pressure set up by the sugar temporarily trapped between the cell wall and plasma membrane. This cell wall damage can be to a large extent averted and the normal morphology and viability retained if the sugar-treated cells are returned to normal media gradually through progressively decreasing concentrations of sugar. We have evidence to suggest that the main function of the sugar in preserving viability on drying lies in its ability to preserve the cell wall intact.

The cell wall, though permeable to mono-, di- and tri-saccharides, is impermeable to proteins and to partially hydrolysed proteins such as constitute a proportion of broth, peptone, etc. By measurements of cell volume we have been able to show that the relatively mild osmotic forces exerted by such substances of higher molecular weight against the cell wall cause its contraction against the plasma membrane and thereby reduce the volume between the two. Thus, when organisms are dried in a mixture of suitable high and low molecular weight components in adequate concentration, the latter will afford protection to the cell wall on drying while the former will limit the amount of the low molecular component inside the cell and thereby reduce the risk of cell wall rupture on reconstitution.

A more detailed account of this work will be published.

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[1] Hellier, G., *J. Bact.*, **41**, 109 (1941).

[2] Fry, R. M., and Greaves, R. I. N., *J. Hygiene*, **49**, 220 (1951).

[3] Powell, E. O., *J. Roy. Micro. Soc.*, **75**, 235 (1956).

GENETICS

Effect of Selection on the Frequency of Genotypes determining Incompatibility in *Prunus avium*

An analysis of the frequency of alleles at the 'S' locus[1] in cultivars of *Prunus avium* indicated that the S_3 allele possessed a selective advantage. Since gametic selection does not appear to operate in this material, and since heterozygosity at the S locus is obligatory, it is of interest to inquire whether selection favours certain genotypes showing inter-allelic interactions, quite apart from the individual alleles they contain. In the absence of genotypic selection, the expected number of varieties of the type S_iS_j is of the form m_im_j. Calculation of an exact value of P for the actual frequencies (Gale, J. S., unpublished work) of the various genotypes is impossibly laborious, but it is possible to set an upper limit to the probability of obtaining by chance a fit as bad as, or worse than, the one observed.

If a_{ij} is the number of varieties of the type S_iS_j, the probability of getting the observed result is:

$$\frac{\dfrac{65\,!}{\pi(a_{ij}!)}\ \pi\left(\dfrac{m_im_j}{65}\right)^{a_{ij}}}{\sum\limits_{\text{all cases}} \dfrac{65\,!}{\pi(a_{ij}!)}\ \pi\left(\dfrac{m_im_j}{65}\right)^{a_{ij}}} = \frac{C}{\sum\limits_{\text{all cases}} C}$$

where $C = \dfrac{1}{\pi(a_{ij}!)}$ and summation is over every way of filling the table, given[1] $a_3 = 42$, $a_1 = 32$, $a_2 = 25$, $a_4 = 15$, $a_5 = 9$ and $a_6 = 7$, where a_i is the observed number of S_i alleles. If C_0 is the value of C for the observed case:

$$P = \frac{\sum\limits_{C \leqslant C_0} C}{\sum\limits_{\text{all cases}} C}$$

and it can be shown (Gale, J. S., unpublished work) that $\sum\limits_{\text{all cases}} C = 337{,}850{,}000\ C_0$ and that the number of cases is 3,693,833, so that $\sum\limits_{C \leqslant C_0} C$ cannot exceed 3,693,833 C_0 and

$$P \leqslant 1{\cdot}0933 \text{ per cent}$$

This upper limit of probability strongly suggests that the observed frequency can be attributed to selection operating on the genotype, and that certain combinations of alleles, in particular $S_1 S_3$, $S_2 S_3$ and $S_1 S_2$, have been favoured in the course of the improvement of the sweet cherry.

Irrespective of whether this effect is due to inter-allelic interactions, or whether more extensive chromosome regions are involved, its significance for plant breeding is obvious. Furthermore, this result clearly demonstrates the response to selection of characters that might otherwise be classified as neutral to the forces operating, and underlines the importance of selection pressure[2] as opposed to genetic drift[3] in determining the frequency of Mendelian characters.

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[1] Williams, W., and Brown, A. G., *Heredity*, **10**, 237 (1956).

[2] Fisher, R. A., and Ford, E. B., *Heredity*, **1**, 143 (1947).

[3] Wright, S., *Evolution*, **2**, 279 (1948).

Chromosomes of South American Amphibians

THE amphibian anurans constitute very interesting material, not only for the cytogenetic study of the evolutionary mechanism of sex determination, but also because they offer prospects from the evolutionary, phylogenetic and cytotaxonomic points of view.

South American species offer a wide scope for investigation due to their abundance and their distribution in different geographical regions of the Continent.

The only species which have been studied up to the present are: *Bufo arenarum* (Hensel)[1] and *Dendrophrynicus stelzneri*[2] which are representatives of the families Bufonidae and Brachycephalidae respectively.

With the view of extending this investigation, we have studied other species belonging to different families (Table 1).

Table 1

Species	Family	Diploid No. ($2n$)	Type of chromosomes
Bufo d'orbignyi	Bufonidae	22	Metacentrics
Odontophrynus americanus	Ceratophrydae	42	Metacentrics
Ceratophrys ornata	Ceratophrydae	88, 92, 98, 108	Metacentrics
Leptodactylus ocellatus	Leptodactylidae	22	Metacentrics
Pseudopaludicola falcipes	Leptodactylidae	22	Metacentrics
Pseudis paradoxa	Pseudidae	22	Metacentrics
Hyla raddiana raddiana	Hylidae	24	Metacentrics

Hyla raddiana raddiana presents, during the first meiotic metaphase, a differential bivalent, that differs from the rest of the autosomes, and is similar to that described in *Bufo arenarum* by one of us[1]. We did not attribute the characteristics of sex chromosomes to their bivalent as Yosida does[3] in a recent paper. The most remarkable fact observed by us is the high number of chromosomes in *Odontophrynus americanus* and *Ceratophrys ornata*.

Odontophrynus americanus has the highest number of chromosomes in an anuran described up to now. *Ceratophrys ornata* is unique among all the amphibians, urodeles and anurans, known up to the present for their high number of chromosomes.

From the cytotaxonomic and evolutionary points of view the taxonomic position of the two genera presents a problem, since they have been considered by Noble[4] as belonging to the family Bufonidae and by Davies[5] to the family Leptodactylidae, the karyotype of which is characterized by a diploid number of $2n = 22$ chromosomes. We suggest that the genera *Odontophrynus* and *Ceratophrys* should be included in a different group, as they appear in the amphibian list published by Cei[6] but forming part of the family Ceratophrydae.

In all the species described here, the chromosomes of the first meiotic metaphase were found to be highly condensed, with two distal localized chiasmata. The primitive annular configuration exhibited by the bivalents during the diplonema remains masked at the moment of the metaphase. None of the species studied shows sex chromosomes that could be differentiated by cytogenetical analysis.

Our observations thus support the views of Galgano[7], Saez *et al.*[8] and Wickbom[9], with reference to the problem of the existence of sex chromosomes in the amphibians.

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[1] Saez, F. A., Rojas, P., and De Robertis, E., *Z. Zellf.*, **24**, 125 (1936).

[2] Saez, F. A., *Physis, Rev. Soc. Arg. Cienc. Nat.*, **18**, 161, 1937 (1939).

[3] Yosida, H. T., *J. Fac. Sci. Hokkaido Imp. Univ.*, **13**, 352 (1957).

[4] Noble, G. K., "The Biology of the Amphibia" (McGraw-Hill Book Co., New York, 1931).

[5] Davies, D. D., *Field Mus. Nat. Hist. Zool.*, **20** (1936).

[6] Cei, J. M., *Inv. Zool. Chilena*, **3**, 31 (1956).

[7] Galgano, M., *Arch. Ital. Anat. Embriol.*, **32**, 171 (1933).

[8] Saez, F. A., Rojas, P., and De Robertis, E., *C.R. Soc. Biol., Paris*, **120**, 368 (1935).

[9] Wickbom, T., *Hereditas*, **31**, 242 (1945).

ANTHROPOLOGY

The Kanam Jaw

SINCE Dr. and Mrs. L. S. B. Leakey discovered that the Oldowan pebble tool-maker in Tanganyika was an Australopithecine (*Zinjanthropus boisei*)[1], I have been frequently asked whether this development has any bearing on the unsolved problem of the Kanam mandible which was recovered by Dr. Leakey in 1932 from deposits in Kenya apparently at the same cultural horizon as the newly discovered Olduvai skull. The Kanam mandible was referred to *Homo kanamensis* sp. nov.[2] and is preserved in the British Museum (Natural History).

As the Kanam mandible has small front teeth and an apparent chin it was regarded by some authorities as indicating the existence of the modern type of man in Lower Pleistocene times, but Prof. P. G. H. Boswell, having formed the opinion in the field that the geological age of the specimen was not beyond doubt, suggested that it should be placed in "a suspense account" pending further discoveries[3].

Attempts have been made during recent years to establish whether the Kanam mandible was or was not contemporaneous with Lower Pleistocene fauna at the site of discovery; but in summary the results were ambivalent. In volcanic soils, particularly under tropical conditions, the chemical dating methods are admittedly unreliable. In 1952 I pointed out that the fluorine/phosphate ratio of the Kanam mandible was higher than that of any fossil bone that had been tested[4]; yet in 1958 radiometric assays (for adsorbed uranium) gave results which led me to write "our doubts about its antiquity have been re-inforced"[5]. But those doubts were primarily morphological in basis.

Prof. P. V. Tobias, of the University of the Witwatersrand, has recently re-investigated the morphology of this mandible with particular reference to the apparent chin or mental eminence (see following communication). Meanwhile, I wish to direct attention to a fact which may have escaped notice. Sir Arthur Keith in 1948 had come to the conclusion that the Kanam mandible was probably Australopithecine. This is what he wrote: "Dr. Leakey believed, and I agree[d] with him, that the Kanam mandible was evidence of the early development of the modern type of man. Both he and I were then ignorant of the fact that small incisors and canines were also characteristic of the South African Dartian anthropoids [Sir Arthur Keith's name for Australopithecines]. It seems to me now to be much more probable that the small front teeth of Kanam man indicate a relationship to Dartian anthropoids rather than to any type of modern man"[6].

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[1] Leakey, L. S. B., *Nature*, **184**, 491 (1959).

[2] Leakey, L. S. B., "Stone Age Races of Kenya", 23 (Oxford Univ. Press, 1935).

[3] *Nature*, **135**, 371 (1935).

[4] Kroeber, A. L., "Anthropology Today", 48 (1952).

[5] Roberts, D. F., and Weiner, J. S., "The Scope of Physical Anthropology", 53 (1958).

[6] Keith, Sir Arthur, "A New Theory of Human Evolution", 261 (1948).

THE controversial mandibular fragment from Kanam in Kenya Colony has long been placed in a 'suspense account' by most palæo-anthropologists. There is little doubt that the presence of an ostensibly well-developed chin in this fossil, for which a Lower Pleistocene age was claimed[1], proved a difficult pill to swallow. As long as the over-simplified notion prevailed that a chin signified *Homo sapiens* and, as long as the Kanam fragment was deemed to possess a chin, one had to explain the unexpected presence of *Homo sapiens* in a Lower Pleistocene deposit or discard either the dating of the deposit or the association of the jaw with the deposit.

In 1955, I was granted permission to re-examine the jaw fragment by the British Museum (Natural History) and, in particular, by Dr. K. P. Oakley and by Dr. L. S. B. Leakey. On first examining the specimen I was struck by two previously unreported facts: first, the 'chin' was represented by an asymmetrical fullness situated largely to the left of the symphyseal surface which had earlier been diagnosed as a "sub-periosteal ossifying sarcoma"[2]. The possibility that the 'chin' was in reality largely a bony reaction to the pathological lesion was heightened by the second observation, namely, that of the elements comprising the mental trigone normally sculptured on the front of a chin, the mental tubercles were completely absent and the mental protuberance was extremely poorly developed. Clearly a detailed morphological re-analysis was essential, and the results of such a study were presented by me at the fourth Pan-African Congress on Prehistory in August 1959[3]. The detailed account will appear in the Proceedings of that Congress.

As a result of the study, I have reached the conclusion that the chin of Kanam man has been greatly over-estimated. In fact, I am convinced that the Kanam fossil does not represent a sapient type of man, but rather belongs to a group of fossils from Africa, known variously as 'palæoanthropic', 'neandertaloid' or 'rhodesioid'. None of the characteristics originally used to define the structure of *Homo kanamensis* (to which the specimen was earlier assigned)[4] is morphologically valid, and the species designation must therefore lapse. On the other hand, a different series of primitive features has impressed me in the re-study. These are:

(1) The great depth in the symphyseal region (40·5 mm. as measured, at least 42·0 mm. as estimated), aligning Kanam with such primitive forms as *Pithecanthropus*, *Atlanthropus*, Mauer and Wadjak, as well as with Cro-Magnon man and several fossils from South and East Africa.

(2) The tremendous depth in the region of M_1 (at least 40·0 mm.), in which respect Kanam exceeds all other mandibles with which it was possible to compare it.

(3) The lack of tapering in body-height between symphyseal and molar regions, which likens the Kanam jaw to those of *Sinanthropus* and *Atlanthropus*, including Sidi Abderrahman.

(4) The very great thickness in the region of M_1 (16·5 mm.), in which feature Kanam is exceeded only by Ternifine I, Heidelberg and Sidi Abderrahman, while it is more or less equal to Ternifine II, *Sinanthropus* G.I., Rabat, Dire-Dawa, Cave of Hearths and La Chapelle.

(5) The rudimentary character of the two elements of the chin present, namely the mentum osseum and the mental protuberance, as in the mandibles of La Quina, La Ferrassie, Spy and Rabat.

(6) The presence of a planum alveolare and of a genial fossa and inferior transverse torus, as in Rabat and Dire-Dawa.

(7) The combination of very small incisors and rather large canines, a likeness with some of the Australopithecinae to which Keith directed attention in 1948, as pointed out by myself at the fourth Pan-African Congress on Prehistory in August 1959 and by Dr. K. P. Oakley (see previous communication).

Several, though not all, of these features might be encountered individually as exceptional variants among modern African mandibles. However, the ensemble of features in a single mandible tends to exclude the Kanam jaw from the *Homo sapiens* group and to relate it rather to a group of earlier African fossil men. Its morphological features align it most closely with the Upper Pleistocene mandible from Dire-Dawa, in the Harrar District of Abyssinia[5], and the Middle Pleistocene Rabat fragment from the

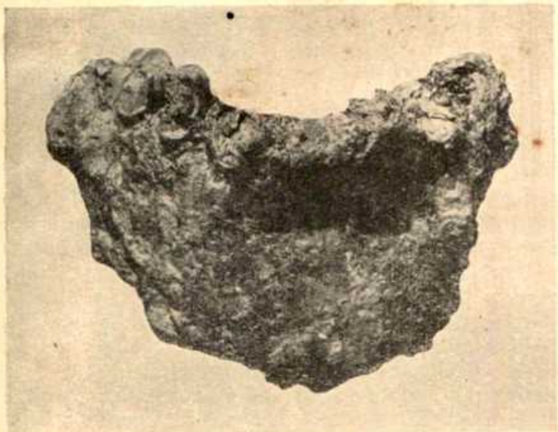

Fig. 1. The Kanam mandible. The fullness of the so-called chin is seen on the right, clearly demarcated by a highlight. The sockets and roots of the missing teeth are apparent; the surviving two right premolar teeth can be seen

Atlantic seaboard of Morocco[6]. On the other hand, the Ternifine mandibles of *Atlanthropus* show more archaic features than do those of Kanam, Rabat and Dire-Dawa; while the Kanam mandible has several morphological features comparable with those of Sidi Abderrahman, though the crucial evidence of the chin region and the retro-symphyseal surface is lacking[7]. As to the possibility of australopithecine affinities, the main resemblance is in the size proportions of the front and back teeth. A few ape-man specimens have, in addition, a degree of chin development faintly reminiscent of that of Kanam.

Thus, while we cannot entirely exclude the possibility that the Kanam jaw is related to a member of the Australopithecinae or of *Atlanthropus*, the balance of evidence supports the view that the Kanam mandible represents a third African fossil showing a cluster of features morphologically intermediate between those of *Atlanthropus* and those of modern man. Whether or not it is justifiable to describe these forms as neandertaloid, thereby relating them by implication to European forms, is a problem which must await more material and further analysis.

At least, it is no longer necessary or correct to build theories upon the alleged 'sapient' or 'modern' morphology of Kanam man; nor should the specimen be interpreted as providing evidence in favour of the *presapiens* theory espoused by Vallois[8]. Perhaps most importantly of all, we are now in a position to consider the age of the Kanam specimen, free from any misconceptions that it is of sapient character.

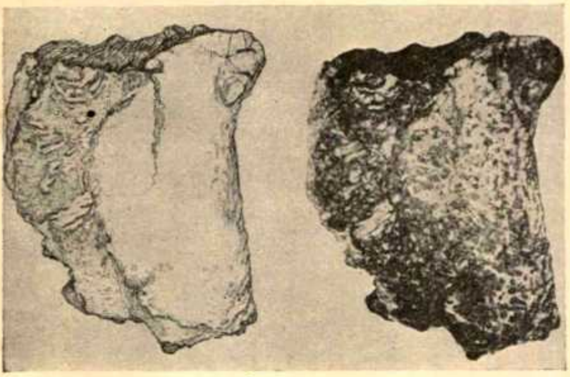

Fig. 2. Sketch and photograph of a section through the Kanam mandible, to show the large pathological mass on the retrosymphyseal surface. The section has passed through the right first incisor root

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[1] Leakey, L. S. B., "Stone Age Races of Kenya" (Oxford Univ. Press, London, 1935).

[2] Lawrence, J. W. P., Appendix *A* in ref. 1, p. 139.

[3] Tobias, P. V., Proc. Fourth Pan-African Congress on Prehistory, Leopoldville (August 1959).

[4] Leakey, L. S. B., *Man*, No. 210, 200 (1933).

[5] Vallois, H. V., *l'Anthropologie*, **55**, 231 (1951).

[6] Vallois, H. V., *C.R. Acad. Sci., Paris*, **221**, 669 (1945).

[7] Arambourg, C., and Biberson, P., *Amer. J. Phys. Anthrop.*, **14**, 467 (1956).

[8] Vallois, H. V., "La Grotte de Fontéchevade. II. Anthropologie", Mem. 29 (Archiv. de l'Inst. de Pal. Hum., 1958).

Fossilized Human Remains from Northern Kenya

DURING the summer of 1959, a party from the Durham Colleges visited the remote district of Turkana in northern Kenya to carry out a geological survey of the Lothidok and the Labur ranges of hills, which border the western shore of Lake Rudolf.

The rocks which compose the Lothidok range are probably Miocene; but a thick sequence of younger lacustrine sediments is developed against the eastern flank of the Lothidok range, immediately south of the Kalalokoil River (approximate position 35° 47′ E., 3° 26′ N.). C. Arambourg[1] has stated that their age is Upper Pleistocene. These lacustrine beds represent deposition in an ancient Lake Rudolf of far greater extent than the existing lake, since they stand between two hundred and three hundred feet above present lake-level. In places, the Miocene lavas and agglomerates of the Lothidok range must have formed a rocky shore-line to this ancient lake, during a part of Pleistocene times. From the western marginal facies of the lake beds, adjacent to their contact with the Miocene, the Durham Colleges party recovered portions of two fossilized human skulls which are the first remains of this kind to be found in the northern region of Kenya. One is represented by little more than the frontal bones and the upper margins of the orbits. The other skull, with which this communication is chiefly concerned, lacks only the face and the left side of the jaws. A few cervical vertebræ and ribs, and part of a humerus were intimately associated with the second skull.

The more complete skull has been slightly distorted during its enclosure in the lake sediments, but it is possible, without further preparation, to distinguish certain salient features of the skull. The cranial bones, which, incidentally, are fully lithified, are unusually thick (12 mm. or more). The skull is long and conspicuously narrow, although the latter character is exaggerated by crushing; and the face appears to have been small. There is no more than a suggestion of a supra-orbital torus, but the brow is remarkably low. The lower jaw (Fig. 1) is very robustly constructed, with a deep horizontal ramus 22 mm. thick below the third molar; and the mandible seems to have possessed a chin. The molar teeth are very badly worn, being planed down almost to the level of the inferior margin of the enamel. They

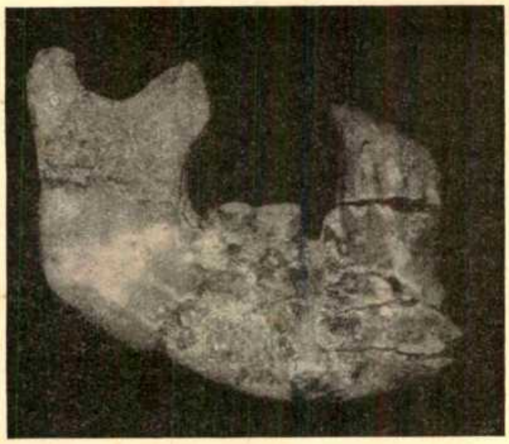

Fig. 1. Lateral aspect of the right branch of a fossilized human mandible, together with a fragment of the upper jaw; from Turkana, northern Kenya (half natural size)

are probably a trifle larger than is the average in Recent individuals of *Homo sapiens*.

Numerous artefacts were found afterwards on sites adjacent to this fossilized human skull, but all were surface finds. None was found *in situ*. The implements include Middle Sangoan hand-axes and Levallois-type flake tools fashioned in chert, small stone points of Solutrean aspect, and barbed harpoon heads and spatulæ of bone. Neighbouring sites also yielded rather poorly preserved remains of various fossilized mammals, including hippos, antelopes and pigs, as yet not properly identified.

It is too early to speculate upon the nature or status of the skull, but there seem to be some clear physical resemblances to the most complete of the Kanjera skulls, which were recovered by L. S. B. Leakey nearly thirty years ago from Pleistocene lake beds developed on the flank of Homa Mountain in western Kenya. Dr. Leakey has suggested a Middle Pleistocene date for the Kanjera skulls. So far, it has not been possible to establish with certainty the precise stratigraphical position of the human fossils from Turkana. Attempts to obtain an absolute dating by chemical methods have also proved inconclusive. The uranium content is the same in both the human bones and in fossilized animal bones from neighbouring sites, which are believed to be undoubtedly contemporaneous with the formation of the lake beds, but the actual figure for all this material is abnormally low.

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[1] Arambourg, C., *Bull. Mus. Hist. Nat., Paris*, 1, fasc. 1 (1935).

PSYCHOLOGY

Age Differences in Movement Pattern

THE literature which describes studies of detailed hand movement patterns contains few references to changes in acceleration during these movements. Such references as do occur are mainly to be found in books on motion study and suggest that a movement occurs in three phases: an acceleration phase to a steady speed, followed by deceleration. For example, Lowry *et al.*[1] give a curve purporting to show this pattern without quoting any evidence to support it. Experimental work carried out here[2] and at Cambridge by Singleton[3] suggested that movement patterns of older people might differ from those of young and, since no relevant evidence could be found in the literature, an ultra high-speed chrono-cyclographic technique was developed in order to make a detailed study of movement patterns.

Our subjects were required to make simple movements in response to a four-choice stimulus and these were photographed with a light on the hand flashing at 100 flashes per second. Three conditions of response were used which differed only in the manipulation demanded after the initial movement was completed. Matched groups of 12 younger subjects (20–25 yr.) and 12 older subjects (60–65 yr.) were used. It was found that under the conditions of this experiment the movement, far from showing a simple acceleration : constant speed : deceleration pattern, showed marked variations in acceleration throughout. Typically, instead of the smooth acceleration which might have been expected, the movement pattern showed very rapid acceleration at the outset, followed by an equally rapid decrease in acceleration to a minimum at approximately 0·1 sec. from the commencement of the movement, which was in turn followed by a second acceleration to a 'peak' of approximately half the value of the first. This whole phase occupied approximately a third of the total movement time. While the characteristic pattern at the outset of the movement was found in both young and old subjects, the old subjects accelerated less rapidly than the young and the 'dip' passed below the zero acceleration line showing that the older men had a short negative acceleration phase. Fig. 1 shows a typical velocity curve for an older subject with the acceleration curve derived from it. There were, within the young and old groups, no statistically significant differences between the three response conditions tested, but between groups there were significant differences in response initiation time in

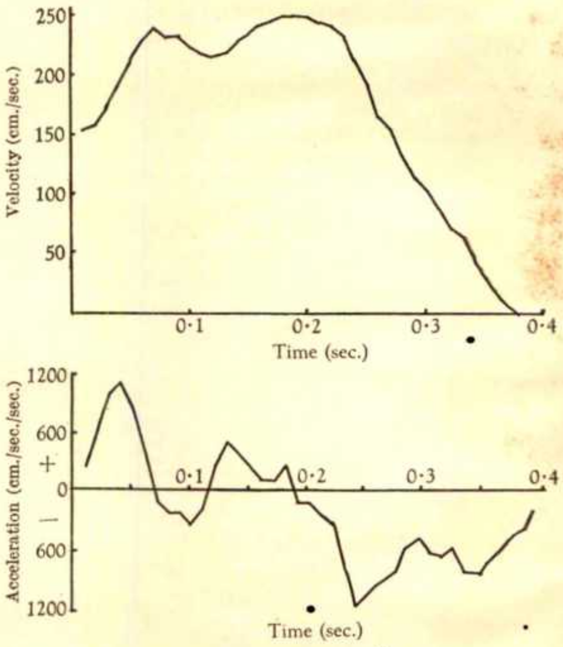

Fig. 1

two of the conditions. Movement times were not significantly different. The magnitude of the initial acceleration and the depth of the 'dip' were significantly different between young and old.

This typical pattern does not appear to occur when a 'free' movement is made in the subject's own time and no stimulus is given or specific response demanded. This suggests that the 'dip' in the initial phase of a 'controlled' movement is in some way associated with either the presence of the visual stimulus or the need to make a response, or both. 'Free' movement patterns do, however, show cyclic periods of acceleration, which are also to be found in the latter two-thirds of the 'controlled' movements. There is a possibility that these may be similar to the oscillations of a particular frequency which have been reported in muscular tremor and static pressure by a number of workers (for example, Hammond *et al.*[4]). If this is so then it would appear that this periodicity will occur in 'free' movements as well as in the static condition.

These results suggest several theoretical possibilities, but the techniques used in this pilot experiment were not sufficiently precise for us to obtain all the information we would need to establish a theoretical model. Work is therefore proceeding with further refined techniques.

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[1] Lowry, S. M., Maynard, H. B., and Stegemerten, G. J., "Time and Motion Study", 129 (McGraw-Hill, New York, 1940).

[2] Griew, S., *Amer. J. Psychol.*, 72, 83 (1959).

[3] Singleton, W. T., *Brit. J. Psychol.*, 14, 166 (1954).

[4] Hammond, P. H., Merton, P. A., and Sutton, G. G., *Brit. Med. Bull.*, 12, 214 (1956).

Experimental Evidence for Extra-Sensory Perception

IN his communication in *Nature* of November 7, p. 1515, Mr. C. E. M. Hansel cites certain results from the Shackleton experiments in precognitive telepathy which he claims support his theory that the guessers made 'card substitutions' on pre-arranged positions on the scoring sheets. In a forthcoming paper in the *Proceedings of the Society for Psychical Research*, I have shown on quite other grounds how extremely improbable it is that such a method was used. I cannot accept Mr. Hansel's arguments based on the 1,139 (+ 2) trials carried out at rapid rate, because, as I shall show, he makes an unwarranted assumption.

What Mr. Hansel has brought to light in his communication is a most interesting 'segmental salience' effect which had previously escaped notice. Card guessers, as a rule, naturally think of a run of 25 trials as being broken up into five successive groups or 'segments' each of five trials. Most scoring sheets have a thick line, drawn after trials 5, 10, 15, 20 and 25. Subconsciously, many guessers tend to envisage their work as a succession of smaller tasks or separate experiments, each consisting of five trials. Now ever since the year 1941, Prof. J. B. Rhine and his colleagues at Duke University have observed that many percipients tend to distribute their 'hits' according to more or less regular patterns throughout the five guesses of a segment, or in the five segments of a run of 25. Three main types of 'salience' have been distinguished: (*a*) 'terminal' salience in which, in the composite pattern of a segment, the first and last trials score above chance expectation while the three middle trials give a score at about 'chance' level or below it; (*b*) 'middle' salience in which the two end trials give scores at, or below, chance expectation and the three middle trials score above 'chance' level; (*c*) 'decline' salience in which, say, the first three trials are well above 'chance' expectation, after which a decline sets in and the score falls to 'chance' level through the last two trials. Similar 'salience' patterns may occur either in the pooled scores of the five trials of the segment or in the five segments of the run, or 'segment' and 'run' salience may occur together, in which case there is sometimes a high degree of positive correlation between them.

These same patterns have been observed during thousands of trials, as, for example, in the long-distance experiments of Dr. Marchesi[1] in which 'middle' salience occurred both in the segment and run over a series of 8,825 trials. It has been noted in 'precognitive' as well as in 'direct' target-guessing.

The Shackleton (+ 2) *trials*. In order to clarify the issue, I shall neglect the 144 trials which occur in positions Nos. 21, 22, 23 of the scoring sheet of 23 (+ 2) trials, since these do not form complete segments. This leaves us with a total of 995 (+ 2) trials in places Nos. 1–20. On these there are 297 (+ 2) hits which are pooled and distributed over the five successive positions in the segment as in Table 1.

Table 1

Position	Trials	Hits	Deviation*	'Odds' against chance
I	199	61	4·2 × S.D.*	> 1·6 × 10^4 to 1
II	200	72	5·6 × S.D.	> 2·6 × 10^7 to 1
III	200	70	5·3 × S.D.	> 1·7 × 10^6 to 1
IV	198	51	2·0 × S.D.	20 to 1
V	198	43	0·6 × S.D.	—
Totals	995	297	—	—

* S.D., standard deviation = $0{\cdot}4\sqrt{n}$, where n is number of trials Expected score, $n/5$.

It is seen that the scoring-rate reaches a peak at Trials II and III, and afterwards declines to the chance-level. It would appear that at the 'rapid' rate of calling, the extra-sensory perception effect is temporarily exhausted after the third trial, and recovers at the start of the next segment (see Fig. 1). On the first three trials there is a total of 203 hits in 599 (+ 2) trials, as compared with a total of 94 hits in 396 (+ 2) trials. The 'salience' in the segment is mainly of type (*C*), and represents a kind of 'position' effect.

So common were these 'salience' effects in the 'segment' or the 'run' that in 1941 Dr. J. A. Greenwood[2] developed a most elaborate statistical method for evaluating 'salience ratios' and they were even employed as a means of detecting the presence of extra-sensory perception in cases where the deviations from chance expectation in an experiment were not in themselves significant[3]. The assumption is that in certain cases extra-sensory perception produces both positive and negative deviations which may cancel each other out in the long run.

By using Fisher's method for evaluating the significance in the difference of the means of two small samples, I find that the odds against the hypothesis that the difference in the scoring-rates of the two groups (I, II, III) and (IV, V) is due to

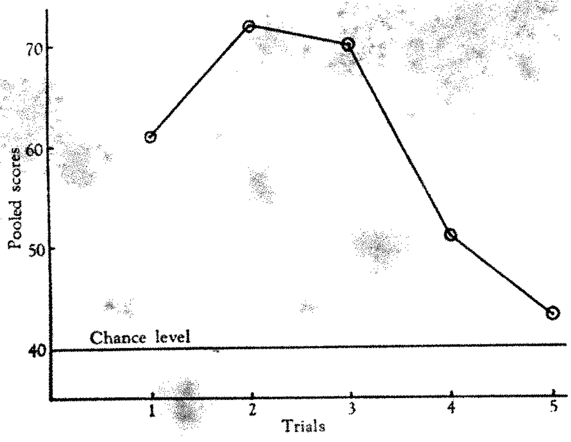

Fig. 1. Score pattern in segment

chance certainly far exceed 1,000 to 1 and are probably of the order of 10^4 to 1, as checked by a different method. The five segments of the run considered as five different units, however, exhibit no evidence of salience; that is, there is not a decline effect in the run itself.

Now the fundamental 'null' hypothesis on which Mr. Hansel has based his expectations of hits and misses in the five trials of the segment is the assumption that, even when extra-sensory perception is functioning at a high level, there ought to be approximately equal numbers of hits (or successes) on equal numbers of trials selected from different pre-assigned positions on the scoring-sheet. But the numerous 'decline' effects observed in both the run and the segment by American investigators show that this assumption is not justified. Further, it has been the experience of Mr. G. N. M. Tyrrell and others that with high scoring subjects such as Miss Johnson, the 'hits' often crowd themselves into unbroken runs of as many as 7 or 8 or more in a run. That is, extra-sensory perception does not function continuously but in spurts. In other words, given a fixed number of hits, these are often found not to be randomly distributed over the 25 cells of the scoring-sheet, but to favour certain positions in the segment or run or in relation to one another.

The 'periodic' effect cited by Mr. Hansel is the result of the 'salience' pattern and constitutes evidence that such a pattern exists that is, on the average, repeated from segment to segment. Similarly, the high scores on the mid-segment trial are simply due to the fact that the peak of the salience happens to coincide with the mid-segment trial and the one preceding.

If an extra-sensory perception salience effect exists, it follows that there may be significantly differing numbers of hits on the different trials of the segment.

Once the existence of extra-sensory perception (the origin of which is utterly mysterious) is admitted, it may be expected to produce many other secondary effects that are surely not more surprising than the existence of the faculty itself.

For, whatever this power may ultimately prove to be, it certainly seems to have some attributes of human intelligence and is not merely a random physical process. It may well be that the subjects who possess this mysterious means of acquiring information have also subconscious personal predilections for certain types of effect, and can at times produce them by unconscious motivation. Thus, we know that one of Prof. Rhines's major subjects could at will orient his mind so as to produce below-chance scores instead of his usual high scores.

S. G. Soal

28 Thurleigh Road,
London, S.W.12.

[1] *J. Parapsychol.*, **6**, 52 (1942).
[2] Greenwood, J. A., *J. Parapsychol.*, **5**, 245 (1941).
[3] Rhine, J. B., *J. Parapsychol.*, **5**, 183 (1941).

Dr. Soal agrees that there is a periodicity in the 'guessers' scores which are related to the line numbers on the score sheet. He calls this effect 'salience' and I will also use this nomenclature. It must, however, be understood that by 'salience' we mean this periodicity in the scores without advancing any theory as to its cause.

Dr. Soal and I agree on the fact that Shackleton consistently produced scores which cannot be attributed to chance. Dr. Soal attributes these scores to telepathy. He regards salience effects as secondary characteristics of telepathy and to explain them he introduces subsidiary hypotheses which can be summarized as follows: (1) Certain guessers 'subconsciously' divide the sequence of twenty-five trials into five 'tasks' of five trials each. Other guessers think of the whole twenty-five trials as the 'task'. (2) At some point in the task, whichever type it be, either towards the end of it, or in the middle of it, or at the two ends of it and not in the middle, some guessers lose their telepathic abilities and their scores fall to the chance-level. (3) In the case of other guessers, they do not lose their telepathic abilities at these positions in the task, but they are "unconsciously motivated" to avoid securing hits and their scores fall below the chance-level.

I have shown that the results of the Soal–Goldney experiment could have been produced by means of a trick which involved memorizing a code. The particular salience effect produced by Shackleton would inevitably arise if such a trick was used, although its presence in the grouped data could be masked if the operators changed their code in a systematic manner.

It is not surprising that salience effects have been observed in other experiments on extra-sensory perception; for they may arise if there are factors which are not adequately controlled in the experimental conditions, or if a trick is being employed. It should further be noted that both errors due to poor experimental design, and confessed trickery, are not uncommon in psychical research, whereas precognition and telepathy are hypothetical processes.

In the case of the Shackleton experiments the issue is quite clear. The scores produced by Shackleton could be obtained by using a trick. Such a trick would, possibly, but not necessarily, reveal itself by causing a periodicity to arise in the scores. Such a periodicity is present in the results of the 'rapid-rate' experiments. It cannot be explained away by calling it 'salience', for this is merely another name for the observed periodicity. To explain salience effects in terms of telepathy, Dr. Soal has produced a number of subsidiary hypotheses which I regard as being as unlikely as is the existence of precognitive-telepathy itself.

C. E. M. Hansel

Department of Psychology,
University of Manchester.

FORTHCOMING EVENTS

*(Meetings marked with an asterisk * are open to the public)*

Monday, March 28

Royal Geographical Society (at 1 Kensington Gore, London, S.W.7), at 5 p.m.—Miss A. Coleman and Mr. K. R. A. Maggs: "A New Land Use Survey".

Radar and Electronics Association (at the Royal Society of Arts, John Adam Street, Adelphi, London, W.C.2), at 7.30 p.m.—Mr. Peter D. Hall: "Computers—Some Design Problems of Large Scale Computers".*

Tuesday, March 29

Institution of Electrical Engineers, Education Discussion Circle (at Savoy Place, London, W.C.2) at 6 p.m.—Discussion on "The Place of Illumination in Electrical Engineering", opened by Mr. G. F. Freeman.

Institution of Mechanical Engineers, Education Group (at 1 Birdcage Walk, Westminster, London, S.W.1), at 6 p.m.—Discussion on "The Need for Post-Graduate Refresher Courses for Engineers in Industry".

Society of Instrument Technology (at Manson House, 26 Portland Place, London, W.1), at 7 p.m.—Mr. G. G. Bloodworth: "Application of Transistors in Instrumentation".

Tuesday, March 29—Thursday, March 31

Institute of Metals (at Church House, Great Smith Street, Westminster, London, S.W.1)—Spring Meeting.

Tuesday, March 29

At 11 a.m.—Sir Ronald Prain, O.B.E.: "Mineral Resources and Metal Reserves" (Presidential Address).

At 6.30 p.m.—Prof. P. M. S. Blackett, F.R.S.: "The Magnetism of Rocks and Continental Drift" (May Lecture).

Wednesday, March 30

Institute of Metals, Metal Physics Committee (in the Hoare Memorial Hall, Church House, Great Smith Street, Westminster, London, S.W.1), at 2.30 p.m.—Meeting on "New Aspects of the Electron Theory of Metals".

Oil and Colour Chemists' Association, London Section (at the Royal Society of Tropical Medicine and Hygiene, Manson House, 26 Portland Place, London, W.1), at 7 p.m.—Dr. H. B. Footner: "Painting in the Petroleum Industry".

Wednesday, March 30—Thursday, March 31

Plastics Institute (at the University of London, London, W.C.1)—Conference on "Polymeric Progress".

Thursday, March 31

Institute of Metals (at Convocation Hall, Church House, Great Smith Street, London, S.W.1), at 10 a.m.—Discussion on "Vacancies, Ageing and Precipitation".

Royal Society (at Burlington House, Piccadilly, London, W.1), at 4.30 p.m.—Dr. N. F. Astbury: "Science in the Ceramic Industry".

Mineralogical Society (at the Geological Society of London, Burlington House, Piccadilly, London, W.1), at 5 p.m.—Scientific Papers.

University of London Institute of Education (in the Great Hall, Goldsmiths' College, New Cross, London, S.E.14), at 5 p.m.—Sir Edward Boyle, Bt., M.P.: "Education—Tradition and Change".*

Institution of Electrical Engineers (at Savoy Place, London, W.C.2), at 5.30 p.m.—Prof. C. F. Powell, F.R.S.: "Cosmic Radiation" (Fifty-first Kelvin Lecture).

APPOINTMENTS VACANT

Applications are invited for the following appointments on or before the dates mentioned:

Lecturer (male) **in Chemistry**—The Principal, Borough Road College, Isleworth, Middlesex (March 31).

Lecturer (Physical Chemist, with postgraduate research experience, and preferably a higher degree and experience in electronics) **in the Department of Electrical Engineering**—Prof. A. Tustin, Department of Electrical Engineering, Imperial College of Science and Technology, London, S.W.7 (March 31).

Lecturer (with a specialized knowledge in at least one of the following fields and prepared to undertake or supervise research: metallurgical analysis, refractories, nonferrous extraction, physical metallurgy) **in Metallurgy**—The Registrar, The Manchester College of Science and Technology, Manchester 1 (April 2).

Research Officer (graduate, with an honours degree in geography, and planning experience) **in the County Planning Department**—The Establishment Officer, Shire Hall, Bedford (April 2).

Biochemist (M.Sc. or Ph.D. in biochemistry or physiological chemistry, and at least three years experience in the procedures of biochemistry or chemical pathology) with the Limerick County Council, Ireland—The Secretary, Local Appointments Commission, 45 Upper O'Connell Street, Dublin (April 5).

Senior Technician (skilled cartographer) **in the Department of Geography**—The Registrar (GST), Queen Mary College (University of London), Mile End Road, London, E.1 (April 8).

Lecturer (medically or non-medically qualified) **in Biochemistry**; an **Assistant** (medically or non-medically qualified) **in Physiology**; and an **Assistant** (preferably applied mathematician) **in Mathematics**—Joint Clerk to the University Court, Queen's College (University of St. Andrews), Dundee (April 9).

Lecturer (with qualifications and a special interest in the teaching of chemistry) **in Education**—The Registrar, The University, Sheffield (April 9).

Assistant Lecturer or **Lecturer** (with a special interest in the philosophy of science) **in Philosophy**—The Registrar, The University, Hull (April 12).

Departmental Assistant in Animal Physiology at the School of Agriculture, Sutton Bonington, near Loughborough, for demonstration and laboratory work in animal physiology, with opportunities for research—The Registrar, The University, Nottingham (April 12).

Lecturer (with special qualifications in engineering geology, including hydrology and soil science) **in the Department of Geology**, University of the Witwatersrand, Johannesburg, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa and London, April 15).

Assistant Professor of Physical Chemistry in the Faculty of Arts and Science, Memorial University of Newfoundland, Canada—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Canada and London, April 15).

University Lecturer in Statistics—The Secretary of the Appointments Committee of the Faculty of Mathematics, the Arts School, Bene't Street, Cambridge (April 18).

Chair of Soil Science in the Faculty of Agriculture, University of Khartoum—The Registrar, University of Khartoum, c/o Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (April 20).

Assistant Lecturers or **Lecturers** (2) (with a good honours degree in geology) **in the Department of Geology**—The Registrar, University College of North Staffordshire, Keele, Staffs (April 23).

Lecturer in the Department of Geography, University of the Witwatersrand, Johannesburg, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa and London, April 25).

Lecturer or **Assistant Lecturer** (man or woman graduate with a special interest in petrology) **in Geology**—The Secretary, Bedford College (University of London), Regent's Park, London, N.W.1 (April 25).

Assistant Lecturer in Physiology at the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, April 29).

Lecturer in Chemical Engineering—The Registrar, University College, Singleton Park, Swansea (April 30).

Post-Doctoral Research Fellow in the Department of Organic Chemistry, to work in collaboration with Dr. A. R. Battersby on the chemistry of natural products, particularly in the alkaloid field—The Registrar, The University, Bristol (April 30).

Scientific Officers/Senior Scientific Officers (with a first- or second-class honours degree, previous research experience in some experimental branch of physics, chemistry or engineering, and at least three years postgraduate experience for Senior Scientific Officer) **in the Basic Physics Division**, National Physical Laboratory, Teddington, Middlesex, to do fundamental research on physical and chemical properties of solids at very high pressures—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting A.119.OA (April 30).

Senior Lecturer in Physical Chemistry at the University of Natal, Pietermaritzburg, South Africa—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (South Africa and London, April 30).

Demonstrator in Physical Chemistry—Head of the Physical Chemistry Laboratory, The University, South Parks Road, Oxford (May 15).

Chief of the Division of Land Research and Regional Survey, Commonwealth Scientific and Industrial Research Organization, Canberra, A.C.T., Australia—Chief Scientific Liaison Officer, Australian Scientific Liaison Office, Africa House, Kingsway, London, W.C.2, quoting Appointment No. 570/84 (May 21).

Post-Doctoral Fellows (2) **in the Chemistry Department**, for work on (*a*) electrode processes and (*b*) thermodynamics of polymer solutions—Prof. B. E. Conway, Department of Chemistry, University of Ottawa, Ottawa, Canada (May 21).

Assistant Editor (man or woman graduate with some knowledge of chemistry, and preferably sub-editing experience with scientific publishers) for the less academic aspects of scientific publications—The General Secretary, The Chemical Society, Burlington House, Piccadilly, London, W.1.

Assistant Experimental Officer (with at least G.C.E., Scottish Leaving Certificate, Higher National Certificate, or equivalent, and some experience of histological techniques), to undertake histological work and to assist in the Pathology Department—The Secretary, Rowett Research Institute, Bucksburn, Aberdeen.

Assistant, Grade B (with a good honours degree, and preferably industrial or research experience), for mathematics—The Clerk to the Governors, South-East Essex Technical College, Longbridge Road, Dagenham, Essex.

Assistant Professor to teach Metallurgical Thermodynamics and Kinetics—Head, Department of Metallurgy, Montana School of Mines, Butte, Montana.

Demonstrator in Biology within the Department of Botany and Zoology—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1, or the Registrar, University of Adelaide, Adelaide, Australia.

Experimental Physicists (with at least a good second-class honours degree and preferably postgraduate experience in some aspect of nuclear physics), to (*a*) collaborate with physicists designing a 7-GeV. proton synchrotron, and take responsibility for aspects of selection, analysis and transport of high energy beams and eventually

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to participate in high energy nuclear research; and (*b*) to work with nuclear physicists using a 50-MeV. linear accelerator and collaborate with university physicists using this machine—The Secretary, Rutherford High Energy Laboratory, Harwell, Didcot, Berkshire, quoting Ref. 1637/34.

MASTER (with high academic qualifications) TO TEACH MATHEMATICS to open scholarship standard; and a MASTER TO TEACH PHYSICS AND/OR CHEMISTRY to A level—The Headmaster, Blundell's School, Tiverton, Devon.

PHYSICIST (with a degree or equivalent in general science including physics, or in chemistry with physics subsidiary, experience in handling radioactive materials and health physics, and preferably a knowledge of electronics and/or general explosive practice) at the Ministry of Aviation H.Q., London, to be responsible for developing effective production methods for radioactive and allied materials and supervision of safety methods allied to health physics at the factory—The Ministry of Labour, Technical and Scientific Register (K), 26 King Street, London, S.W.1, quoting Ref. A.118/O.A.

PRINCIPAL GRADE PHYSICIST (with experience either in hospital physics or in some relevant field, such as biophysics, electronics or nuclear physics), to initiate work on physics applied to the various fields of medicine (other than radiotherapy)—The House Governor, The Queen Elizabeth Hospital, Edgbaston, Birmingham 15.

SENIOR LABORATORY TECHNICIAN, to be responsible for new physics teaching laboratories and lecture demonstrations up to degree standard—The Secretary, King's College, Strand, London, W.C.2, quoting Ref. 3066.

SENIOR TECHNICIAN II (with some organizing ability, and preferably previous experience in medical or biological research) IN THE CANCER RESEARCH LABORATORY, for work in connexion with the cytology and cytochemistry of human tumours—The Administrative Officer, Mount Vernon Hospital, Northwood, Middx.

SOIL CHEMIST (with an honours degree in chemistry, or in natural science including chemistry, and at least two years postgraduate training or experience as a soil chemist) at the West African Institute for Oil Palm Research, Nigeria, for the study and analysis of soil types with a view to improving soil conditions—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.197/199/016/T.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

British Steel Castings Research Association. British and Foreign Specifications for Steel Castings, Part 1—Summaries, Comparisons and other relevant Information. Pp. xiv+48. (Sheffield: British Steel Castings Research Association, 1959.) 40*s.*; 7.50 dollars. [121

Society for Freedom in Science. Occasional Pamphlet No. 19 (December, 1959): Opposition Aroused in China by Marxist Planning of Scientific Research. Pp. ii+16. (Oxford: Dr. John R. Baker, Dept. of Zoology, University Museum, 1959.) [121

Ministry of Agriculture, Fisheries and Food. Fishery Investigations, Series II, Vol. XXII, No. 8: The Spawning of the Plaice (*Pleuronectes platessa*) in the Irish Sea. By A. C. Simpson. Pp. ii+30. (London: H.M. Stationery Office, 1959.) 9*s.* net. [121

University Scholarships for Student Apprentices in Electrical, Electronic and Mechanical Engineering. Pp. vii+23 (7 plates). (London: H.M. Postmaster General, 1960.) [121

Institute of Physics. The Teaching of Mathematics to Physicists. Revised Report. Pp. 25. (London: Institute of Physics, 1959.) 2*s.* 6*d.* [121

Ministry of Labour. Factories Acts, 1937 and 1948. Factories-(Ionising Radiations)-Special Regulations—Second Preliminary Draft of Regulations. Pp. 20. (London: H.M. Stationery Office, 1960.) 1*s.* 3*d.* net. [121

Society for the Bibliography of Natural History. Essai sur la Geographie des Plantes (1807). Par Alexander von Humboldt. (Sherborn Fund Facsimile, No. 1). Pp. 38+1 plate. (London: Society for the Bibliography of Natural History, c/o British Museum (Natural History), 1959.) 10*s.* [121

At the Farmer's Service: a Handy Reference to Various Services available to Farmers in England and Wales. Pp. 79. (London: Ministry of Agriculture, Fisheries and Food, 1959.) [121

University of Cambridge School of Agriculture Memoirs. No. 31: A summary of the papers published by the members of the Staff of the School of Agriculture and its Associated Research Organisations during the period Oct. 1st, 1958—Sept. 30th, 1959. Review Series No. 14: Statistics—a Retrospect. Pp. 28. (Cambridge: University of Cambridge School of Agriculture, 1959.) [121

The Registrar General's Statistical Review of England and Wales for the year 1958. Part 2: Tables, Population. Pp. xi+181. (London: H.M. Stationery Office, 1960.) 11*s.* 6*d.* net. [261

Ministry of Housing and Local Government: Central Advisory Water Committee. Final Report of the Trade Effluents Sub-Committee of the Central Advisory Water Committee. Pp. iv+52. (London: H.M. Stationery Office, 1960.) 3*s.* 6*d.* net. [261

Ciba (A.R.L.), Ltd. Technical Notes, No. 205 (January 1960): The Durability of 'Aerolite' 300. Pp. 20. (Duxford: Ciba (A.R.L.), Ltd., 1960.) [261

Southern Regional Council for Further Education. Twelfth Annual Report, October 1959. Pp. 40. (Reading: Southern Regional Council for Further Education, 1959.) [261

Memoirs of the Royal Astronomical Society. Vol. 68, Part 2: A Survey of Radio Sources at a Frequency of 159 Mc/s. By D. O. Edge, J. R. Shakeshaft, W. B. McAdam, J. E. Baldwin and S. Archer. The Positions, Flux Densities and Angular Diameters of 64 Radio Sources Observed at a Frequency of 178 Mc/s. By B. Elsmore, M. Ryle and Patricia R. R. Leslie. Pp. 37–67. (London: Royal Astronomical Society, 1959.) 12*s.*; 1.90 dollars. [261

Royal Institute of Chemistry. Lectures, Monographs and Reports, 1959. No. 3: Some Chemical Aspects of Plant Disease Control. By Dr. R. L. Wain. Pp. ii+21. 4*s.* 6*d.* No. 4: Chemical Mechanisms in the Induction of Cancer. By Dr. Alexander Haddow. Pp. ii+21. 4*s.* 6*d.* net. No. 5: Modern Explosives. By the late Dr. Wilfrid Taylor. Pp. 63. 6*s.* net. (London: Royal Institute of Chemistry, 1959.) [261

G.B. Film Library. Educational Films, Issue No. 26. Pp. 120. Filmstrips, 1959–1960. Pp. 72. (Perivale, Middx.: G.B. Film Library, 1959.) [261

Other Countries

University of California Publications in Geological Sciences. Vol. 36, No. 2: Pliocene and Lower Pleistocene of the Western Part of the San Francisco Peninsula. By William Glen. Pp. 147–198+plates 15–17. (Berkeley and Los Angeles: University of California Press: London: Cambridge University Press, 1959.) 1 dollar. [61

Annual Report of the Board of Regents of the Smithsonian Institution showing the Operations, Expenditures, and Condition of the Institution for the year ended June 30, 1958. (Publication No. 4354.) Pp. x+559+107 plates. (Washington, D.C.: Government Printing Office, 1959.) 3.75 dollars. [61

National Institute of Genetics, Japan. Annual Report, No. 9, 1958. Pp. v+144. (Misima, Sizouka-ken: National Institute of Genetics, 1959.) [61

The Plant Galls of Norway: a Preliminary Catalogue based on the collection of the Zoological Museum, University of Bergen. By Donald Leatherdale. (Universitetet i Bergen Årbok 1959—Naturvitenskapelig rekke Nr. 8.) Pp. 56. (Bergen: A. S. John Griegs Boktrykkeri, 1959.)

Centro Brasileiro de Pesquisas Fisicas. Notas de Fisica. Vol. 5, No. 10: Algebras of Finite Differential Order and the Operational Calculus. By Leopoldo Nachbin. Pp. 105–142. Vol. 5, No. 11: The Form Factors in $K\mu$, and Ke Decay. By S. W. MacDowell. Pp. 143–155. Vol. 5, No. 12: Gamma Rays in the Decay of 141[illegible]a. By E. W. Cybulska and L. Marquez. Pp. 157–163. Vol. 5, No. 13: Decay of $\Lambda_0 \rightarrow n + \gamma$ Through Non Local Fermi Interactions Due to Vector Mesons. By J. L. Acioli. Pp. 165–177. (Rio de Janeiro: Centro Brasileiro de Pesquisas Fisicas, 1959.) [61

India: Council of Scientific and Industrial Research. National Metallurgical Laboratory Annual Report, 1958–59. Pp. iii+49. (Jamshedpur, Bihar: National Metallurgical Laboratory, 1959.) [121

Proceedings of the American Philosophical Society. Vol. 103, No. 6 (December 15, 1959): Studies of Historical Documents in the Library of the American Philosophical Society. Pp. ii+727–821. (Philadelphia: American Philosophical Society, 1959.) 1 dollar. [121

Journal of Nuclear Energy, Part C: Plasma Physics—Accelerators—Thermonuclear Research, Vol. 1, Nos. 1/2 (October, 1959). Pp. 104. Subscription rates (a) per volume: 20 dollars, 140*s.* including postage. (b) per annum for subscribers who notify the Publisher direct certifying that the journal is for their private use, 30 dollars, 210*s.* including postage. Back numbers 28 dollars, 200*s.* per volume. (New York and London: Pergamon Press, 1959.) [121

Companhia de Diamantes de Angola (DIAMANG). Servicos Culturais. Dundo—Lunda—Angola. Museu do Dundo. Subsidios para o Estudo da Biologia na Lunda. Publicacoes Culturais No. 42: Contribution à l'Étude de la Flore de la Lunda d'Après les Récoltes de Gossweiler (1946–1948). Par A. Cavaco. Pp. 230. (Lisboa: Companhia de Diamantes de Angola, 1959.) [261

University of California Publications in Botany. Vol. 31, No. 1: Cytogenetic and Evolutionary Studies in the Genus *Dactylis*. 1: Morphology, Distribution, and Interrelationships of the Diploid Subspecies. By G. Ledyard Stebbins and D. Zohary. Pp. 1–40+plates 1–3. (Berkeley and Los Angeles: University of California Press; London: Cambridge University Press, 1959.) 75 cents. [261

Metropolitan Life Insurance Company. Statistical Bulletin, Vol. 40 (November–December 1959): New Weight Standards for Men and Women. Fathers are Younger. How Fatal Accidents Occur in the Home. Geographic Variations in Mortality by Cause. Pp. 12. (New York: Metropolitan Life Insurance Company, 1959.) [261

Mitteilungen aus der Biologischen Bundesanstalt für Land- und Forstwirtschaft, Berlin-Dahlem. Heft 97 (November 1959): 32 Deutsche Pflanzenschutz-Tagung der Biologischen Bundesanstalt für Land- und Forstwirtschaft in Hannover, 6-10 Oktober 1958. Pp. xxii+239. (Berlin-Dahlem: Biologische Bundesanstalt für Land- und Forstwirtschaft, 1959.) 12.80 D.M. [261

Bernice P. Bishop Museum Special Publication No. 47: Fishhooks. By Kenneth P. Emory, William J. Bonk and Y. H. Sinoto. (Hawaiian Archaeology). Pp. xiii+45+6 plates. (Honolulu: Bernice P. Bishop Museum, 1959.) [261

Commonwealth of Australia. Department of Supply—Australian Defence Scientific Service. Annual Report of the Defence Standards Laboratories for the year ended 30th June, 1959. Pp. iv+75. (Maribyrnong, Victoria: Defence Standards Laboratories, 1959.) [261

Queensland. Fourteenth Annual Report of the Council of the Queensland Institute of Medical Research, year ended 30th June, 1959. Pp. 24. (Brisbane: Queensland Institute of Medical Research, 1959.) [261

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